Janssen Research & Development *

Clinical Protocol

A Randomized, Observer-blind, First-in-Human Phase 1/2a Study to Evaluate the Safety, Reactogenicity and Immunogenicity of Three Different Doses of VAC52416 (ExPEC10V) in Adults Aged 60 to 85 Years in Stable Health

Protocol VAC52416BAC1001; Phase 1/2a
Amendment 8

VAC52416 ExPEC10V

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

PROTOCOL AMENDMENT SUMMARY OF CHANGES TABLE

DOCUMENT HISTORY	
Document	**Date**
Amendment 8	19 October 2021
Amendment 7	18 March 2021
Amendment 6	24 November 2020
Amendment 5	15 July 2020
COVID-19 Appendix	14 May 2020 (incorporated in the protocol as of Amendment 7)
Amendment 4	17 March 2020
Amendment 3	09 October 2019
Amendment 2	05 April 2019
Amendment 1	21 January 2019
Original Protocol	04 December 2018

Amendment 8 (19 October 2021)

Overall Rationale for the Amendment: To shorten the long-term follow-up period in Cohort 2 from 3 years to 1 year, as a 3-year long-term follow-up period will be part of another Phase 3 ExPEC study that includes the same population, ie, participants with a history of UTI. The long-term follow-up period in Cohort 1 will be extended from 3 years to 5 years, to obtain more long-term data on the safety and immunogenicity of the selected dose of the ExPEC10V vaccine in the general population. In addition, the assessment of vaccine-induced functional antibodies against the ExPEC10V O8 serotype will be removed from the MOPA analysis in Cohort 1 and 2, except for baseline and Day 15 in Cohort 1, as the assay was not able to detect vaccine-induced functional antibodies against the O8 serotype. Clarifications about the Cohort 1 and 2 LTFU procedures will be added, and minor changes will be implemented to align text in this protocol with recent template updates.

Section Number and Name	Description of Change	Brief Rationale
1.1 Synopsis 1.2 Schema 1.3.3 Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants) 3.2 Cohort 2 - Double-Blind Period With Double-Blind Long-Term Follow-up Period (N=±420) 4.1.2 Cohort 2 4.4.2.2 Long-Term Follow-Up Period 8 STUDY ASSESSMENTS AND PROCEDURES 8.4 Immunogenicity Assessments 9.5.2 Cohort 2	Shortened the long-term follow-up period of Cohort 2 from 3 years to 1 year by removing Year 2 (Day 731) and Year 3 (Day 1096).	This decision was made because a 3-year long-term follow-up period will be part of another Phase 3 ExPEC study that includes the same population, ie, participants with a history of UTI.
1.1 Synopsis 1.2 Schema 1.3.3 Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind	Extended the long-term follow-up period of Cohort 1 from 3 years to 5 years by adding Year 4 (Day 1461) and Year 5 (Day 1826).	This decision was made to obtain more long-term data on the safety and immunogenicity of the selected dose of the ExPEC10V vaccine in the general population.

Section Number and Name	Description of Change	Brief Rationale
Long-Term Follow-up Period (All Participants) 3.1 Cohort 1 - Phase 1/2a Observer-Blind Period With Open-Label Long-Term Follow-up Period (N=404) 4.1.1 Cohort 1 4.1.2 Cohort 2 4.4.1.2 Long-Term Follow-Up Period 4.4.2.2 Long-Term Follow-Up Period 8 STUDY ASSESSMENTS AND PROCEDURES 8.4 Immunogenicity Assessments 9.5.1 Cohort 1		
1.3.3 Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants)	Clarified that participants in Cohort 1 who do not give consent for the Year 4 and Year 5 LTFU visits will be seen as early withdrawal.	This decision was made because it allows for the most relevant data analysis and handling after implementation of this amendment.
1.3.3 Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants) 8 STUDY ASSESSMENTS AND PROCEDURES	Clarified that participants in Cohort 2 who have already completed their Year 1 LTFU visit will receive a safety follow-up telephone call, explaining they have completed the study due to an amendment to the protocol.	To inform participants as soon as possible after implementation of this amendment, since they have completed the study and are not yet aware of it.
8.2.5 Pregnancy	Deleted optional sponsor template text indicating that follow-up information regarding any postnatal sequelae in the infant will be retrieved, if possible.	To be consistent with the company process.
8.4 Immunogenicity Assessments	Removed the assessment of vaccine-induced functional antibodies against the ExPEC10V O8 serotype from the MOPA analysis for participants from Cohort 1 and 2, except for baseline and Day 15 in Cohort 1.	This decision was made because the assay was not able to detect vaccine induced functional antibodies against the O8 serotype.
10.2 Appendix 2: Regulatory, Ethical, and Study Oversight Considerations	Added text regarding protocol clarification communication.	To align the protocol text with the sponsor's current protocol template.

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1. PROTOCOL SUMMARY

1.1. Synopsis

A Randomized, Observer-blind, First-in-Human Phase 1/2a Study to Evaluate the Safety, Reactogenicity and Immunogenicity of Three Different Doses of VAC52416 (ExPEC10V) in Adults Aged 60 to 85 Years in Stable Health.

ExPEC10V (primary compound number: VAC52416; JNJ-69968054) is a 10-valent vaccine candidate in development for the prevention of invasive extraintestinal pathogenic *Escherichia coli* (ExPEC) disease (IED) in adults 60 years of age and older. ExPEC10V consists of the O-antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the carrier protein, a genetically detoxified form of exotoxin A (EPA) derived from *Pseudomonas aeruginosa*.

Invasive ExPEC disease (IED) is defined as an acute illness consistent with systemic bacterial infection that is microbiologically confirmed either by the isolation and identification of *E. coli* from blood or other normally sterile body sites, or by the isolation and identification of *E. coli* from urine in a patient with signs and symptoms of invasive disease (presence of systemic inflammatory response syndrome [SIRS], sepsis or septic shock) and no other identifiable source of infection.

OBJECTIVES AND ENDPOINTS

This study's aim is to assess the safety, reactogenicity, and immunogenicity of 3 different doses of ExPEC10V and to select the optimal dose for further clinical development (Cohort 1). Cohort 2 is aimed to expand the dataset supporting the short- and long-term safety and immunogenicity of the optimal dose of ExPEC10V, selected from the primary analysis results of Cohort 1. Cohort 2 will include approximately 420 male and female participants in stable health with a history of urinary tract infection (UTI) in the past 5 years and will be included in the study to support the plan for late stage development of ExPEC10V.

COHORT 1 - Phase 1/2a observer-blind period with open-label long-term follow-up period (N=404):

Objectives	Endpoints
Primary	
• To evaluate the safety and reactogenicity of different doses of ExPEC10V in participants ≥60 to ≤85 years of age	• Solicited local and systemic adverse events (AEs) collected for 14 days post-vaccination (from Day 1 to Day 15) • Unsolicited AEs collected from the administration of the study vaccine until 29 days post-vaccination (from Day 1 to Day 30) • Serious adverse events (SAEs) collected from the administration of the study vaccine until Day 181
• To evaluate the dose-dependent immunogenicity of ExPEC10V on Day 15 in participants ≥60 to ≤85 years of age	• Antibody titers for ExPEC10V, as determined by multiplex electrochemiluminescent (ECL)-based immunoassay and multiplex opsonophagocytic assay (MOPA) on Day 15
Secondary	
• To evaluate the correlation between multiplex ECL-based immunoassay (total antibody) and MOPA (functional antibody) serum titers on Day 15	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 15
• To evaluate the dose-dependent immunogenicity of ExPEC10V on Days 30 and 181 in participants ≥60 to ≤85 years of age	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Days 30 and 181
• To evaluate, in the long-term follow-up (LTFU) period, the safety of the ExPEC10V dose selected for further clinical development based on the Day 30 primary analysis in participants ≥60 to ≤85 years of age	• SAEs related to the study vaccine or study procedures collected from Day 182 until the end of the study
• To evaluate, in the LTFU period, the immunogenicity of the ExPEC10V dose selected for further clinical development based on the Day 30 primary analysis	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826)

COHORT 2 - Double-blind period with double-blind long-term follow-up period (N=±420):

Objectives	Endpoints
Primary	
• To evaluate the safety and reactogenicity of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• Solicited local and systemic AEs collected for 14 days post-vaccination (from Day 1 to Day 15) • Unsolicited AEs collected from the administration of the study vaccine until 29 days post-vaccination (from Day 1 to Day 30) • SAEs collected from the administration of the study vaccine until Day 181
• To evaluate the immunogenicity of the selected dose of ExPEC10V on Day 30 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 30
Secondary	
• To evaluate the correlation between multiplex ECL-based immunoassay (total antibody) and MOPA (functional antibody) serum titers on Day 30 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 30
• To evaluate the immunogenicity of the selected dose of ExPEC10V on Days 15 and 181 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay on Days 15 and 181 and MOPA on Day181
• To evaluate, in the LTFU period, the safety of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• SAEs related to the study vaccine or study procedures collected from Day 182 until the end of the study
• To evaluate, in the LTFU period, the immunogenicity of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA at Year 1 (Day 366)

Objectives	Endpoints
Exploratory	
• To evaluate the effect of ExPEC10V on the intestinal (stool) microbiome by metagenomic analyses	• Metagenomics of stool samples from a selected subset[a] of participants to evaluate the effect of ExPEC10V on: Prevalence of pathogens (eg, *Clostridium difficile*) in the intestinal flora Prevalence of ExPEC10V serotypes in the intestinal flora

Hypothesis

No formal hypothesis testing will be done. One of the three ExPEC10V doses tested in Cohort 1 will be selected for further clinical development based on the safety (through Day 30) and immunogenicity data (Day 15 results obtained by multiplex ECL-based immunoassay and MOPA). The dose selection will consider the totality of the evidence available at the time of the primary analysis (Day 30) for Cohort 1, including an immunogenicity dose-selection algorithm that will guide the decision.

In Cohort 2, the safety and immunogenicity of the ExPEC10V dose selected from Cohort 1 will be further evaluated in participants in stable health with a history of UTI in the past 5 years.

OVERALL DESIGN

This is a Phase 1/2a randomized, multicenter, interventional study including two cohorts.

For Cohort 1, the study will have an observer-blind, active-controlled design, and a total of 404 adult participants aged ≥60 to ≤85 years in stable health with or without a history of UTI will be included. The study design for Cohort 1 is comprised of three periods: a maximum 28-day screening period, an observer-blind 181-day follow-up period with vaccination on Day 1 and an open-label LTFU period which will last from Day 182 until 5 years (Day 1826) post-vaccination (Figure 1). Only participants from the ExPEC10V selected dose group (approximately 100 participants) and participants from the Prevnar 13 group (approximately 52 participants) will progress to the LTFU period. The end of Cohort 1 will be the last participant's Year 5 visit (Day 1826) in Cohort 1.

For Cohort 2, the study will have a double-blind, placebo-controlled design, and a total of approximately 420 adult participants aged ≥60 years in stable health with a history of UTI in the past 5 years will be included. Enrollment will commence after completion of the Phase 1/2a primary analysis and ExPEC10V dose selection from Cohort 1. The study design for Cohort 2 is comprised of three periods: a maximum 28-day screening period, a double-blind 181-day follow-up period with vaccination on Day 1, and a double-blind LTFU period which will last from Day 182 until 1 year (Day 366) post-vaccination (Figure 2). All participants in Cohort 2 will progress to the LTFU period. The end of Cohort 2 will be the last participant's Year 1 (Day 366) visit in Cohort 2.

The end of study will be the last participant's Year 5 visit (Day 1826) in Cohort 1.

[a] A subset of 33% of participants selected at randomization on Day 1 (using the interactive web response system [IWRS]).

Cohort 1: Phase 1

In Phase 1 of Cohort 1, a total of 84 participants will be enrolled in a staggered approach following stepwise dose-escalating procedures with safety evaluations in place before progressing from one step to the next. An internal Data Review Committee (DRC) will be commissioned for this study to review the physical examination data (baseline as well as targeted), baseline demographic data and the 14-day post-vaccination safety data (including solicited local and systemic AEs, unsolicited AEs, SAEs, clinical laboratory data and vital signs) of these 84 Phase 1 participants.

In this phase of the study, participants will be enrolled and randomized in six steps:

- **Step 1**: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V low dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups.
- **Step 2**: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V low dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups.
- **Step 3**: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V medium dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups.
- **Step 4**: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V medium dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups.
- **Step 5**: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V high dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups.
- **Step 6**: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V high dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups.

All participants will receive a single intramuscular (IM) injection of either ExPEC10V (1 of 3 doses), ExPEC4V or Prevnar 13 on Day 1 per the assigned study vaccination groups. The four sentinel participants at each of Steps 1, 3 and 5 will be contacted by telephone 24 hours post-vaccination to collect safety information. The blinded 24-hour post-vaccination safety data in each group of four sentinel participants will be reviewed by the principal investigator (PI), study responsible physician (SRP) and sponsor medical lead (SML). Randomization of additional participants for the next step will be halted until this Day 2 sentinel safety evaluation is completed (Figure 3).

In the absence of any clinically significant findings, upon decision by the PI, SRP and SML, an additional 24 participants (for Steps 2, 4 and 6) will be enrolled and randomized to one of three study vaccination groups (Table 1, Figure 3) to receive a single IM injection of either ExPEC10V (low dose in Step 2, medium dose in Step 4 and high dose in Step 6), ExPEC4V or Prevnar 13 on Day 1.

After vaccination of an additional 24 participants at each dose level (low dose in Step 2, medium dose in Step 4, and high dose in Step 6), 14-day post-vaccination safety data of all 28 (4+24) participants at each dose level will be reviewed by the DRC before progressing to the next dose level or Phase 2a. Further randomization will be halted until the DRC safety evaluation is completed at each step (Figure 3). The DRC will review blinded data first but may also review unblinded data, if deemed necessary.

Cohort 1: Phase 2a

Based on acceptable safety and reactogenicity (in the absence of any safety concerns or any events meeting a specific study vaccination pausing rule) as determined by the DRC after the review of 14-day post-vaccination safety data for the initial 84 participants, the remaining 320 participants from Cohort 1 will be randomized and dosed in Phase 2a. These additional 320 participants will be enrolled and randomized in parallel in a ratio of 2:2:2:1:1 to one of the five study vaccination groups to receive a single IM injection of either ExPEC10V (1 of 3 doses), ExPEC4V or Prevnar 13 on Day 1 (Table 1).

In addition to performing the 14-day safety review for the initial 84 participants, the DRC will also evaluate safety data of Cohort 1 over the course of the study and review any events that meet a specific study vaccination pausing rule or any other safety issue that may arise. The DRC will review blinded data first but may also review unblinded data, if deemed necessary.

For Cohort 1, the primary analysis will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. The final analysis will occur when all participants have completed the Day 181 visit or have discontinued earlier. The study sites will be informed after the final analysis database lock which participants should progress to the LTFU period (ExPEC10V selected dose group and Prevnar 13 group) and for which participants the Day 181 visit will be the last on-site visit. For all participants, a telephone contact will be scheduled to inform them whether they will be progressing to the LTFU period. For participants not progressing to the LTFU period, that telephone contact will be their last study visit.

For participants progressing to the open-label LTFU period, yearly follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to the time of the visit at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826) after vaccination.

A participant in Cohort 1 will be considered to have completed the observer-blind period of the study if he or she has completed Day 181. For participants in Cohort 1 progressing to the open-label LTFU period of the study, a participant will be considered to have completed the study if he or she has completed the open-label LTFU period (Year 5 visit, Day 1826).

Cohort 2

In Cohort 2, the safety, reactogenicity, and immunogenicity of the selected dose of ExPEC10V (based on the primary analysis results of Cohort 1) will be evaluated in participants aged ≥60 years in stable health with a history of UTI in the past 5 years. For Cohort 2, the study will have a double-blind, placebo-controlled design, and a total of approximately 420 participants will be enrolled and randomized in parallel in a 2:1 ratio (280 participants in the ExPEC10V group and 140 in the placebo group).

All participants will receive a single IM injection of either the selected dose of ExPEC10V or placebo on Day 1 per the assigned study vaccination groups (Table 2). If a specific study vaccination pausing rule has been met or if any other safety issue may arise, the DRC will be convened to evaluate safety data of Cohort 2. The DRC will review blinded data first but may also review unblinded data, if deemed necessary.

For Cohort 2, the primary analysis will include safety and immunogenicity data and will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. The final analysis will occur when all participants have completed the Day 181 visit or have discontinued earlier. For all participants, follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to the time of the visit at Year 1 (Day 366) after vaccination.

A participant in Cohort 2 will be considered to have completed the study if he or she has completed the double-blind LTFU period (Year 1 visit, Day 366).

A stool sample analysis will be performed in a selected subset of participants to evaluate the effect of ExPEC10V on the prevalence of pathogens (eg, *Clostridium difficile*) and ExPEC10V serotypes in the intestinal flora using metagenomics.

NUMBER OF PARTICIPANTS

A total of approximately 824 participants will be enrolled in the study; 404 participants in Cohort 1 and approximately 420 participants in Cohort 2.

VACCINATION GROUPS AND DURATION

Description of Vaccinations

ExPEC10V (VAC52416): *E. coli* bioconjugate vaccine in phosphate buffered solution containing O-antigen PSs of ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the EPA carrier protein.

Single 0.5 mL IM (deltoid) injection of one of the three doses of ExPEC10V on Day 1.

ExPEC4V (JNJ-63871860): *E. coli* bioconjugate vaccine in saline buffer solution containing O-antigen PSs of ExPEC serotypes O1A, O2, O6A, and O25B (4:4:4:8 µg PS/ExPEC serotypes) separately bioconjugated to the EPA carrier protein. ExPEC4V dose was identified from the primary analysis data of the Phase 2 study 63871860BAC2001.

Single 0.5 mL IM (deltoid) injection of ExPEC4V on Day 1.

Prevnar 13: Sterile suspension of saccharides of the capsular antigens of *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F, individually linked to non-toxic diphtheria CRM197 protein.

Single 0.5 mL IM (deltoid) injection of Prevnar 13 on Day 1, supplied in a single-dose prefilled syringe.

Placebo: Normal saline.

Single 0.5 mL IM (deltoid) injection of placebo on Day 1.

In Cohort 1, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation and administration. Study vaccine will be administered IM by an unblinded vaccine administrator at the study site who can be a trained and qualified study nurse, medical doctor, or otherwise qualified healthcare professional. These unblinded members will not be part of the team performing the evaluations.

In Cohort 2, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation. These unblinded members will not be part of the team performing the evaluations. The vaccine administrator will be blinded and can perform other study evaluations.

SAFETY EVALUATIONS

Key safety assessments include solicited local and systemic AEs, unsolicited AEs, SAEs, physical examinations, vital sign measurements, and, for Cohort 1 only, clinical laboratory tests.

IMMUNOGENICITY EVALUATIONS

For all participants, key immunogenicity assessments of collected sera will include the assessment of ExPEC10V and ExPEC4V serotype-specific total immunoglobulin G antibody levels elicited by the vaccine as measured by multiplex ECL-based immunoassay, and ExPEC10V and ExPEC4V serotype-specific functional antibodies as measured by MOPA. Immunogenicity assessments of pneumococcal antibody titers elicited by Prevnar 13 will not be performed.

OTHER EVALUATIONS

For Cohort 2, stool samples will be analyzed using metagenomics to evaluate the effect of ExPEC10V on the prevalence of pathogens (eg, *Clostridium difficile*) and ExPEC10V serotypes in the intestinal flora.

STATISTICAL METHODS

For Cohort 1, the primary objective of the study is to evaluate the safety and reactogenicity of different doses of ExPEC10V. The probability of observing at least 1 AE occurring at a rate of 1/100 is 63% with 100 participants receiving ExPEC10V per dose group. It was estimated that if no (S)AE will be observed in a dose group (N=100), this would provide 95% confidence that the true incidence is no more than 2.95%.

For Cohort 1, the co-primary objective of the study is to evaluate the dose-dependent immunogenicity of ExPEC10V, as measured by multiplex ECL-based immunoassay and MOPA. Based on the geometric mean ratio (GMR), in order to have 90% power to get a significant result (one-sided alpha=5%) if two groups are differing 2-fold (0.301 on $\log_{10}$-scale) for a certain O-serotype, assuming a standard deviation (SD)=0.7 (which was the case for O25B), 94 participants per ExPEC10V dose group will be needed. And with a 5% dropout rate, 100 participants per ExPEC10V dose group will be a reasonable sample size for Cohort 1 of the study.

For Cohort 2, the primary objective of the study is to evaluate the safety and reactogenicity of the selected dose of ExPEC10V. The probability of observing at least 1 AE occurring at a rate of 1/100 is 94% with 280 participants receiving ExPEC10V. At a rate of 1/1,000, the probability of observing at least 1 AE is 24%. It was estimated that if no (S)AE will be observed in the safety cohort (Cohort 2) ExPEC10V group (N=280), this would provide 95% confidence that the true incidence is no more than 1.1%.

Safety Analyses

No formal statistical testing of safety data is planned. The safety analysis will include the descriptive summary (including 95% confidence intervals) of solicited local AEs, solicited systemic AEs, unsolicited AEs, and SAEs. The overall frequencies per study vaccination group as well as frequencies according to severity and duration will be calculated for solicited and unsolicited AEs.

Summaries, listings, datasets, or participant narratives may be provided, as appropriate, for those participants who die, who discontinue study vaccination due to an AE, or who experience a severe AE or an SAE. For Cohort 1, a listing of participants with any laboratory results outside the reference ranges will be provided.

Descriptive statistics of temperature, pulse/heart rate, respiratory rate, and blood pressure (systolic and diastolic) (supine) values and changes from baseline will be summarized at each scheduled time point.

Immunogenicity Analyses

For ExPEC10V serotype antibodies as measured by multiplex ECL-based immunoassay and MOPA, and EPA as measured by multiplex ECL-based immunoassay only, the following measures of immunogenicity will be evaluated and tabulated by the study vaccination groups, for all immunogenicity time points:

- proportion of participants with a ≥2-fold and ≥4-fold increase in serum antibody titers from Day 1 (pre-vaccination)

- geometric mean titer (GMT)
- GMR: fold change from baseline, calculated from the post-baseline/baseline value.

For the LTFU period, descriptive summaries of immunogenicity will be presented for each serotype.

Immunogenicity Dose-Selection Algorithm (Cohort 1)

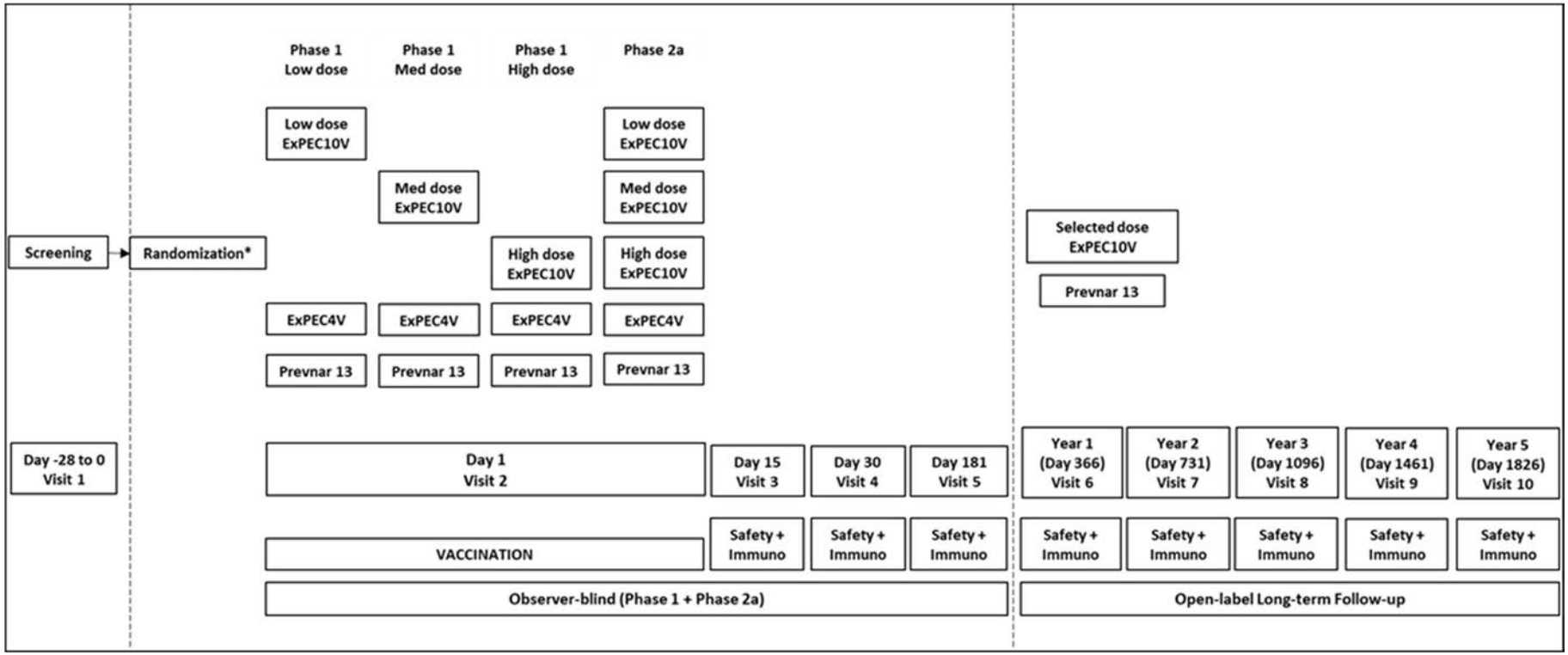
The dose selection will consider the totality of the evidence available at the time of the primary analysis for Cohort 1, including an immunogenicity dose-selection algorithm that will guide the decision.

The immunogenicity dose-selection algorithm will include the $\log_{10}$ transformation of the fold increase from baseline to Day 15 as the response variable and the independent variables will include the dose groups and the $\log_{10}$ transformations of the baseline titer.

The algorithm is a stepwise procedure where in the first four steps, the $\log_{10}$ transformation of the fold increase from baseline to Day 15 for the serotypes O25B, O6A, O2, and O1A (in that order) are included in the model as a response variable. If after these four steps, more than one group is retained (see below), the other six serotypes are included in the model as a response variable and the dose group that is selected the most will be used as the selected dose group. In each step, both the least squares mean of each of the dose groups included in the model and a 95% confidence interval of the difference between the least squares mean of a dose group and that dose group having the highest least squares mean are computed. The dose group having the highest least squares mean and those other dose groups that are non-inferior to this dose group based on the confidence limits are retained. Only these dose groups are included in the model as covariate in the next step. The dose selected by the immunogenicity dose-selection algorithm will be the dose group retained after the final step.

1.2. Schema

Figure 1: Cohort 1: Overall Study Design



Immuno Immunogenicity; Med Medium

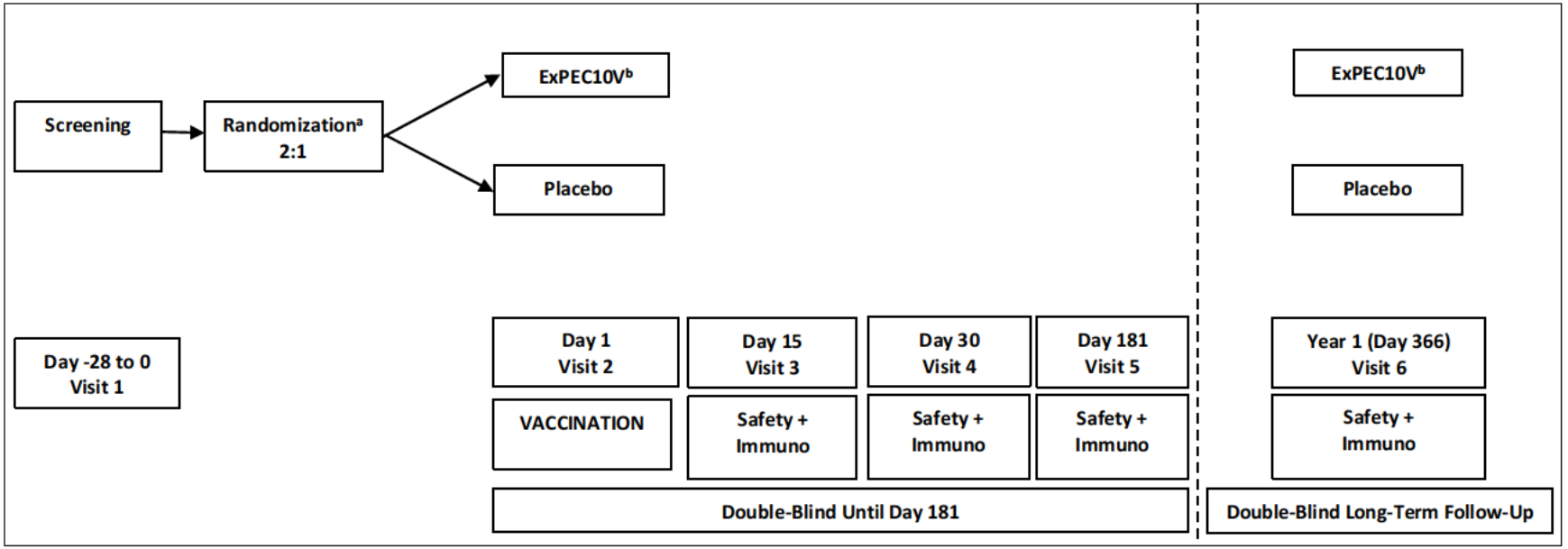
* For randomization schedule, refer to Table 1.

If any participant experiences any issue with the electronic diary (ediary) entry at any time in between vaccination (Day 1) and 14 days post vaccination (Day 15) visit, an optional telephone contact(s) should be made to collect safety data.

In addition, all participants will be contacted by telephone 2 days post vaccination (Day 3) to collect safety information and after the final analysis database lock to inform all participants whether that telephone contact will be their last study visit or if they will be progressing to the long term follow up (LTFU) period.

The dose selection for ExPEC10V will be based on the primary analysis (Day 30) results.

Figure 2: Cohort 2: Overall Study Design



Immuno Immunogenicity
[a] For randomization schedule, refer to Table 2.
[b] The ExPEC10V dose used in Cohort 2 will be based on the primary analysis (Day 30) results of Cohort 1.
If any participant experiences any issue with the electronic diary (ediary) entry at any time in between vaccination (Day 1) and 14 days post vaccination (Day 15) visit, an optional telephone contact(s) should be made to collect safety data. In addition, all participants will be contacted by telephone 2 days post vaccination (Day 3) to collect safety information.
Stool samples will be collected from all participants on Day 1 and from a subset of 33% randomly selected participants on Days 30 and 181 (refer to Section 1.3.2).

Table 1: Cohort 1: Vaccination Schedule

		Phase 1						Phase 2a	Total
		Step 1	Step 2	Step 3	Step 4	Step 5	Step 6	Step 7	
Study Vaccination Group	**Vaccination on Day 1**	**Sentinel participants (Low dose)**	**Additional participants (Low dose)**	**Sentinel participants (Medium dose)**	**Additional participants (Medium dose)**	**Sentinel participants (High dose)**	**Additional participants (High dose)**	**Additional Phase 2a Participants**	
G1	Low dose ExPEC10V*	2	18					80	100
G2	Medium dose ExPEC10V*			2	18			80	100
G3	High dose ExPEC10V*					2	18	80	100
G4	ExPEC4V**	1	3	1	3	1	3	40	52
G5	Prevnar 13***	1	3	1	3	1	3	40	52
Total		**4**	**24**	**4**	**24**	**4**	**24**	**320**	**404**

G Group

* ExPEC10V consists of the O antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the carrier protein, a genetically detoxified form of exotoxin A (EPA) derived from *Pseudomonas aeruginosa*.

** ExPEC4V consists of the O antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O6A, and O25B separately bioconjugated to the carrier protein, a genetically detoxified form of exotoxin A (EPA) derived from *Pseudomonas aeruginosa*.

*** Prevnar 13, Pneumococcal 13 valent conjugate vaccine (diphtheria CRM197 protein) is a sterile suspension of saccharides of the capsular antigens of *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F, individually linked to non toxic diphtheria CRM197 protein.

The randomization ratio for the sentinel participants will be 2:1:1 (Steps 1, 3 and 5) and randomization ratio for the additional participants in Phase 1 will be 6:1:1 (Steps 2, 4 and 6).

The 24 hour post vaccination safety in sentinel participants will be monitored and reviewed by principal investigator (PI), study responsible physician (SRP) and sponsor medical lead (SML). Randomization of additional participants will be halted until this Day 2 sentinel safety evaluation is completed.

After dosing of 28 participants at each dose level, a Data Review Committee (DRC) review will be performed to evaluate the 14 day post vaccination safety data before progressing to the next dose level or Phase 2a. (The double vertical lines indicate a DRC review)

The randomization ratio for the Phase 2a participants will be 2:2:2:1:1 (Step 7).

Total Phase 2a participants (404) include the 84 Phase 1 participants and additional 320 participants.

Table 2: Cohort 2: Vaccination Schedule

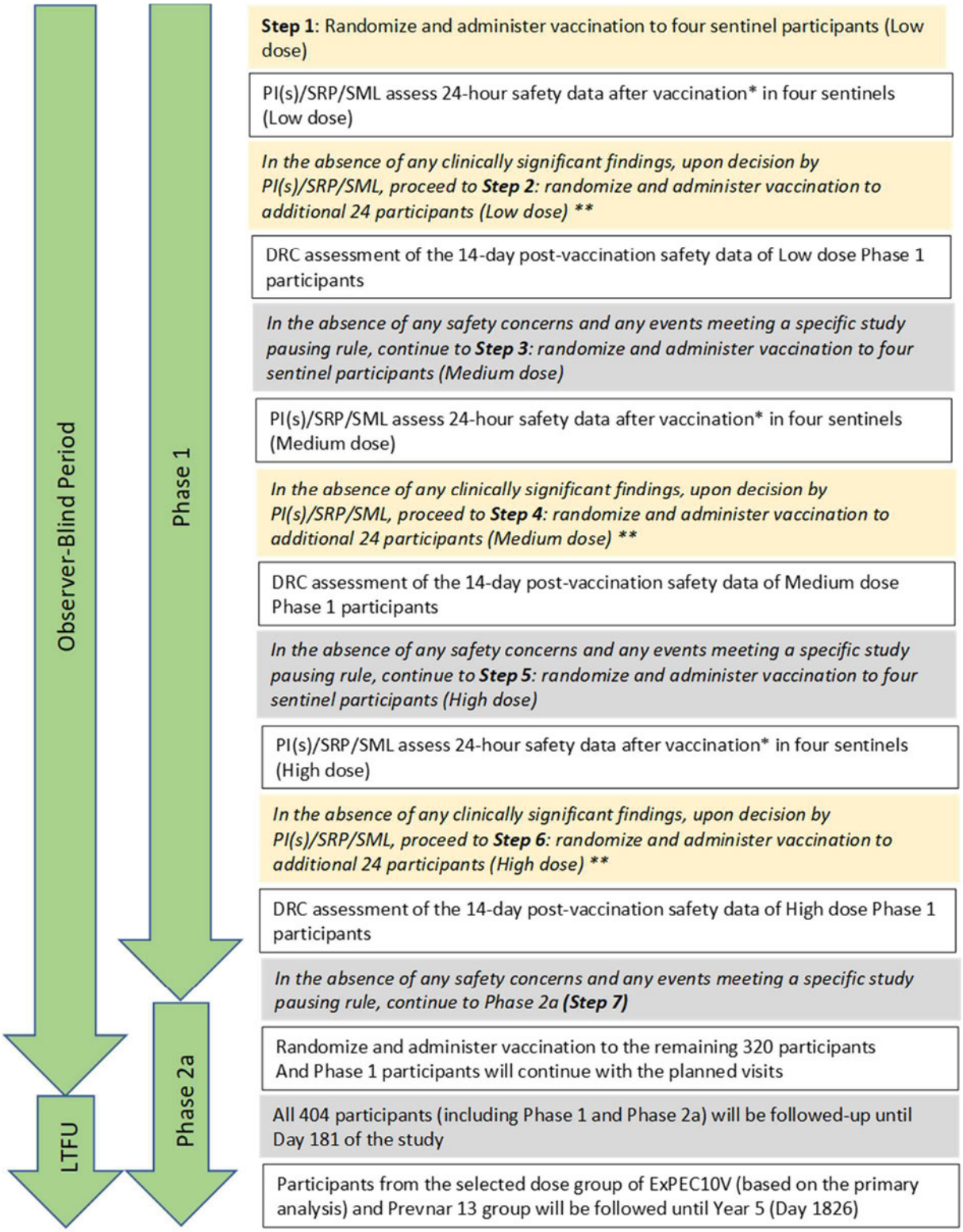
Study Vaccination Group	Vaccination on Day 1	Total[b]
G6	ExPEC10V[a]	280
G7	Placebo	140
Total		**420**

G Group

[a] ExPEC10V consists of the O antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B, and O75 separately bioconjugated to the carrier protein, a genetically detoxified form of exotoxin A (EPA) derived from *Pseudomonas aeruginosa*.
The randomization ratio for the participants enrolled in Cohort 2 of the study will be 2:1 (ExPEC10V:Placebo). The ExPEC10V dose used in Cohort 2 will be based on the primary analysis (Day 30) results of Cohort 1.

[b] The total number of participants to be included in each study vaccination group is an approximation.

Figure 3: Cohort 1: Participant Enrollment and Vaccination Scheme



DRC data review committee; LTFU long term follow up; PI principal investigator; SML sponsor medical lead; SRP study responsible physician.

* Sentinel participants will be contacted by telephone 24 hours post vaccination to collect safety information.

** Participants per dose level (28 participants for each dose level and 84 participants in total)

1.3. Schedule of Activities (SoA)

1.3.1. Cohort 1: Observer-Blind Period

Clinic Visit #	1	2	☎	☎	3	4	5	☎	
Visit Timing		VACC	VACC +1 d	VACC +2 d	VACC +14 d	VACC +29 d	VACC +6 mo		Early Exit [a]
Visit Day	-28 to 0	1	2	3	15	30	181		
Visit Window			0	+1 d	±2 d	±3 d	±14 d		
Visit Type	Screening	VACCINATION [c]	Telephone contact: only for sentinel participants	Telephone contact: Safety	Safety and Immuno	Safety and Immuno	Safety and Immuno	Telephone contact: For LTFU [b]	Early exit
Written informed consent [d]	●								
Inclusion/exclusion criteria	●	❶							
Demographics	●								
Medical history/prestudy meds [e]	●	❶							
Physical examination including body weight and height	●	❶ ❷			❷	❷	❷		❷
Vital signs [f] including body temperature	●	❸			●	●	●		●
12 lead ECG [g]	●								
Serology (HIV 1/2, hepatitis B/C)	●								
Randomization [h]		❶							
Pre vaccination symptoms [i]		❶							
Safety laboratory blood sample (in mL)	● 15	❶ 5			● 5	● 5			❼ 5
Immunogenicity blood sample (in mL)		❶ 10			● 10	❺ 10+50	● 10		● 10
Vaccination		●							
30 minutes post vaccination observation [j]		●							
Participant ediary distribution [k]		●							
Participant ediary review by site staff [l]					●	❻			❹
Solicited AE recording		●	●	●	●				❹
Unsolicited AE recording [m]		●	●	●	●	●			❼
SAE recording [m]		●	●	●	●	●	●		●
Concomitant medications [n]		●	●	●	●	●	●		●

AE adverse event; d day(s); DRC data review committee; ECG electrocardiogram; eCRF electronic case report form; ediary electronic diary; HIV human immunodeficiency virus; ICF informed consent form; LTFU long term follow up; meds medications; mo month; SAE serious adverse event; VACC vaccination

☎ telephone contact; ❶ pre vaccination; ❷ targeted physical examination including body weight, ❸ pre and post vaccination; ❹ if within 14 days of the vaccination; ❺ an additional immunogenicity blood sample of 50 mL will be collected from a total of 30 participants (up to 15 participants in low and medium dose groups, up to 15 participants in high dose group and if needed, remainder from Phase 2a) willing to donate blood; ❻ if Visit 3 occurs before 14 days post vaccination; ❼ if within 29 days of the vaccination

a. An early exit visit will be conducted for the participants who are unable to continue participation in the study and withdraw from the study before Day 181, but who do not withdraw consent.
b. The study sites will be informed after the final analysis database lock which participants should progress to LTFU period (ExPEC10V selected dose group and Prevnar 13 group; see LTFU period Schedule of Activities in Section 1.3.3) and for which participants the Day 181 visit will be the last on site visit. For all participants, a telephone contact will be scheduled to inform them whether that telephone contact will be their last study visit or whether they will be progressing to the LTFU period.

c. Enrollment and vaccination of participants will be done in a staggered approach following the stepwise dose escalation procedures to allow for safety review of four sentinel participants at 24 hours after vaccination followed by a 14 day post vaccination safety data review of 28 participants by DRC at each dose level (first at low dose, second at medium dose, followed by high dose). Sentinel participants will be contacted by telephone 24 hours post vaccination to collect safety information.
d. Signing of the ICF should be done before any study related activity.
e. Prestudy specific therapies (non steroidal anti inflammatory drugs, corticosteroids, antihistaminic, and vaccinations) administered up to 30 days before signing the ICF must be recorded at screening for all participants.
f. Vital signs (systolic and diastolic supine blood pressure, respiratory rate and pulse/heart rate) including body temperature will be measured at screening, before vaccination and at the end of the 30 minute observation period after vaccination (measurements should be done after 5 minutes of seated rest).
g. Supine ECG after at least 5 minutes rest, and prior to vital signs measurements and blood draw.
h. Randomization to either of the five study vaccination groups comprised of three groups of ExPEC10V (low, medium or high dose), one group of ExPEC4V, and one group of Prevnar 13 on Day 1.
i. Investigator must check for acute illness or oral temperature ≥38.0°C (100.4°F) at the time of vaccination. In such cases, the participant may be vaccinated up to, and no later than 28 days within the screening window, or be withdrawn at the discretion of the investigator.
j. Participants will be closely observed for a minimum of 30 minutes post vaccination. Any unsolicited AEs, solicited local and systemic AEs, and vital signs (supine systolic and diastolic blood pressure, pulse/heart rate respiratory rate and oral temperature) will be documented by study site personnel following this observation period.
k. At Visit 2, participants will be provided with an ediary (to record solicited local and systemic AEs), a thermometer (to measure body temperature), and a ruler (to measure diameter of any erythema and swelling), and will be instructed to measure and record solicited local and systemic AEs and body temperature daily for 14 days post vaccination (day of vaccination and the subsequent 14 days). The participants will also be handed wallet cards.
   Optional phone call(s) to participants to collect safety data, only if participant experiences issues with ediary entry at any time in between Visit 2 (Day 1) and Visit 3 (Day 15).
l. If Visit 3 occurs before the end of the diary period after the vaccination, review of the ediary will still take place, but the ediary will be returned by the participant at the next study visit (Day 30).
   ***Note**: If any of the first 84 participants come in earlier than Day 15 for Visit 3 (allowed window is ±2 days), a subsequent phone call will be made at the end of the diary period to collect ediary information recorded between the actual visit and the end of the diary period on Day 15, which will be captured in the eCRF for DRC review.*
m. All AEs and special reporting situations, whether serious or non serious, that are related to study procedures or that are related to non investigational (concomitant) Janssen products will be reported from the time a signed and dated ICF is obtained onwards. All other AEs, SAEs and special reporting situations, will be reported from the day of first vaccination onwards. SAEs will be collected from vaccination until Day 181 in the study.
n. Concomitant therapies will be collected from the signing of the ICF until 29 days after the vaccination, and additionally outside of these periods when associated with an SAE until Day 181.

1.3.2. Cohort 2: Double-Blind Period Until Day 181

Clinic Visit #	1 [a]	2 [a]	☎	3	4	5	
Visit Timing		VACC	VACC +2 d	VACC +14 d	VACC +29 d	VACC +6 mo	Early Exit [b]
Visit Day	-28 to 1 [a, c]	1	3	15	30	181 [d]	
Visit Window			+1 d	±2 d	±3 d	±14 d	
Visit Type	Screening	VACCINATION	Telephone contact: Safety	Safety and Immuno	Safety and Immuno	Safety and Immuno	Early exit
Written informed consent [e]	●						
Inclusion/exclusion criteria	●	❶					
Demographics	●						
Medical history/prestudy meds [f]	●	❶					
Physical examination including body weight and height	●	❶ ❷		❷	❷	❷	❷
Vital signs [g] including body temperature	●	❸		●	●	●	●
Stool samples for intestinal flora analysis [h]		❶			●	●	
Randomization [i]		❶					
Pre vaccination symptoms [j]		❶					
Immunogenicity blood sample (in mL)		❶ 10		● 10	● 10	● 10	● 10
Vaccination		●					
30 minutes post vaccination observation [k]		●					
Participant ediary distribution [l]		●					
Participant ediary review by site staff [m]				●	❺		❹
Solicited AE recording		●	●	●			❹
Unsolicited AE recording [n]		●	●	●	●		❻
SAE recording [n]		●	●	●	●	●	●
Concomitant medications [o]		●	●	●	●	●	●

AE adverse event; d day(s); eCRF electronic case report form; ediary electronic diary; ICF informed consent form; LTFU long term follow up; meds medications; mo month; SAE serious adverse event; VACC vaccination

☎ telephone contact; ❶ pre vaccination; ❷ targeted physical examination including body weight, ❸ pre and post vaccination; ❹ if within 14 days of the vaccination; ❺ if Visit 3 occurs before 14 days post vaccination; ❻ if within 29 days of the vaccination

a. Screening will be performed within 28 days prior to vaccination or on the day of vaccination. If screening is performed on the day of vaccination, Visit 1 and Visit 2 will coincide on Day 1.
b. An early exit visit will be conducted for the participants who are unable to continue participation in the study and withdraw from the study before Day 181, but who do not withdraw consent.
c. Screening must be completed and all eligibility criteria must be fulfilled prior to randomization and vaccination.
d. All participants will progress to the LTFU period.
e. Signing of the ICF should be done before any study related activity.
f. Prestudy specific therapies (non steroidal anti inflammatory drugs, corticosteroids, antihistaminic, and vaccinations) administered up to 30 days before signing the ICF must be recorded at screening for all participants.
g. Vital signs (systolic and diastolic supine blood pressure, respiratory rate and pulse/heart rate) including body temperature will be measured at screening, before vaccination, and at the end of the 30 minute observation period after vaccination (measurements should be done after 5 minutes of seated rest).
h. All participants will be provided with a stool sample kit at the screening visit to collect a baseline stool sample, which should be available before vaccination. Baseline stool samples should be collected at home by each participant between 1 and 7 days prior to the scheduled Day 1 visit; stool samples should be provided to the study site personnel upon arrival on Day 1. At randomization on Day 1, a subset of 33% of participants will be randomly selected (using the interactive web response system [IWRS]) to provide stool samples at the Day 30 and 181 visits. The selected subset of participants will be provided with a stool sample kit at the visits preceding these stool sampling visits. Stool samples for the Day 30 and Day 181 study visits should be collected at home by the participant between 1 and 7 days prior to the respective study visits and provided to the study site personnel upon arrival for these visits. For the Day 30 and Day 181 visits only, if the

participant does not bring the stool sample to the visit, he or she may deliver the stool sample to the study site up to 3 days after the visit.

i. Randomization to either ExPEC10V or placebo on Day 1.
j. Investigator must check for acute illness or oral temperature ≥38.0°C (100.4°F), or ongoing or suspected symptomatic UTI at the time of vaccination. In such cases, the participant may be vaccinated up to, and no later than 28 days within the screening window, or be withdrawn at the discretion of the investigator.
k. Participants will be closely observed for a minimum of 30 minutes post vaccination. Any unsolicited AEs, solicited local and systemic AEs, and vital signs (supine systolic and diastolic blood pressure, pulse/heart rate, respiratory rate, and oral temperature) will be documented by study site personnel following this observation period.
l. At Visit 2, participants will be provided with an ediary (to record solicited local and systemic AEs), a thermometer (to measure body temperature), and a ruler (to measure diameter of any erythema and swelling), and will be instructed to measure and record solicited local and systemic AEs and body temperature daily for 14 days post vaccination (day of vaccination and the subsequent 14 days). The participants will also be handed wallet cards. Optional phone call(s) to participants to collect safety data, only if participant experiences issues with ediary entry at any time in between Visit 2 (Day 1) and Visit 3 (Day 15).
m. If Visit 3 occurs before the end of the diary period after the vaccination, review of the ediary will still take place, but the ediary will be returned by the participant at the next study visit (Day 30).
n. All AEs and special reporting situations, whether serious or non serious, that are related to study procedures or that are related to non investigational (concomitant) Janssen products will be reported from the time a signed and dated ICF is obtained onwards. All other AEs, SAEs, and special reporting situations will be reported from the day of first vaccination onwards. SAEs will be collected from vaccination until Day 181 in the study.
o. Concomitant therapies will be collected from the signing of the ICF until 29 days after the vaccination, and additionally outside of these periods when associated with an SAE until Day 181. Use of oral licensed *E. coli* vaccines (eg, Uro Vaxom [OM 89] or Uromune) is allowed for the duration of the study, including long term follow up. Vaccination with an authorized/licensed Coronavirus Disease 2019 (COVID 19) vaccine (eg, vaccination for COVID 19 through Emergency Use Authorization) is permitted when given at least 28 days before or after planned administration of the study vaccination. Use of *E. coli* or COVID 19 vaccines must always be recorded as concomitant therapy.

1.3.3. Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants)

Clinic Visit #	**6**[f]	**7**[e]	**8**[e]	**9**[e]	**10**[e]		
Visit Timing	**VACC + 1 Year**	**VACC + 2 Years**	**VACC + 3 Years**	**VACC + 4 Years**	**VACC + 5 Years**	**Early Exit**[a]	**End of study call**[i]
Visit Day	**Day 366**	**Day 731**	**Day 1096**	**Day 1461**	**Day 1826**		
Visit Window	**±14 d**	**±28 d**	**±28 d**	**±28 d**	**±28 d**		
Visit Type	**Safety and Immuno**	**Safety and Immuno**	**Safety and Immuno**	**Safety and Immuno**	**Safety and Immuno**		
Informed Consent before entering LTFU (only applicable for Cohort 1)	●			●[g, h]	●[g, h]		
Immunogenicity blood sample (in mL)	● 10	● 10	● 10	● 10	● 10	●[b] 10	
SAE recording [c]	●	●	●	●	●	●	●
Concomitant medications [d]	●	●	●	●	●	●	

d day(s); Immuno Immunogenicity; LTFU long term follow up; SAE serious adverse event; VACC vaccination

a. Participants who are unable to continue participation in the study, but who do not withdraw consent, should be encouraged to complete an exit visit, at the investigator's discretion. If the exit visit would occur shortly after an annual visit, the investigator may decide to have the exit visit replaced by a telephone contact.
b. Not applicable if the exit visit is replaced by a telephone contact.
c. Only serious adverse events (SAEs) related to study vaccine or study procedures will be collected during the LTFU period.
d. Only medications in conjunction with SAEs related to the study vaccine or study procedures should be recorded. Use of oral licensed *E. coli* vaccines (eg, Uro Vaxom [OM 89] or Uromune) is allowed for the duration of the study, including long term follow up. Vaccination with an authorized/licensed COVID 19 vaccine (eg, vaccination for COVID 19 through Emergency Use Authorization) is permitted when given at least 28 days before or after planned administration of the study vaccination. Use of *E. coli* or COVID 19 vaccines must always be recorded as concomitant therapy.
e. The Year 2 (Day 731), Year 3 (1096), Year 4 (Day 1461), and Year 5 (Day 1826) LTFU visits are applicable to Cohort 1 only.

f. The Year 1 (Day 366) LTFU visit applies to both Cohort 1 and Cohort 2.
g. Participants in Cohort 1 will need to provide their reconsent for the Year 4 and Year 5 safety and immunogenicity LTFU visits, the latest just before the Year 4 visit.
h. Participants in Cohort 1 who do not provide their reconsent for the Year 4 and Year 5 LTFU visits will be seen as early withdrawal. A participant in Cohort 1 will be considered to have completed the study if he or she has completed the open label LTFU period (Year 5, Day 1826).
i. Participants in Cohort 2 who have already completed their Year 1 LTFU visit at the time of implementation of protocol amendment 8, will receive a safety follow up telephone call, informing them they have completed the study due to the implementation of a protocol amendment. This telephone call should be performed as soon as possible, and before the canceled Year 2 LTFU visit that was planned to be performed under the original protocol, after implementation of protocol amendment 8.

2. INTRODUCTION

ExPEC10V (primary compound number: VAC52416; JNJ-69968054) is a 10-valent vaccine candidate in development for the prevention of invasive extraintestinal pathogenic *Escherichia coli* (ExPEC) disease (IED) in adults 60 years of age and older. ExPEC10V consists of the O-antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the carrier protein, a genetically detoxified form of exotoxin A (EPA) derived from *Pseudomonas aeruginosa*.

ExPEC are a leading and rising cause of bacteremia and bloodstream infections worldwide, and comprise 17% to 37% of clinically significant blood isolates.[15,18,22] In the United States (US), the overall annual incidence of ExPEC bloodstream infections in adults ranges between 30 and 50/100,000 person-years.[22,28] In Europe, the number of cases of ExPEC bacteremia annually increased by 8.1% between 2002 and 2008.[15] A retrospective 15-year evaluation of bloodstream infections in Denmark (1992-2006) also showed an increasing incidence of gram-negative bloodstream infections, with the majority of community-onset (40%-50%), hospital-acquired (10%-25%) and healthcare-associated (30%-40%) bacteremia caused by ExPEC, and mostly in the older adults >60 years of age.[27] A recent study found an annual incidence of 63.5/100,000 person-years in England (2013-2014).[13] The worldwide emergence of multidrug resistance (ie, resistance to three or more antibiotic classes) among ExPEC strains, such as *E. coli* sequence type 131:O25B, represents a major challenge for the prevention and management of ExPEC infections.[24,25]

Invasive ExPEC disease (IED) is defined as an acute illness consistent with systemic bacterial infection that is microbiologically confirmed either by the isolation and identification of *E. coli* from blood or other normally sterile body sites, or by the isolation and identification of *E. coli* from urine in a patient with signs and symptoms of invasive disease (presence of systemic inflammatory response syndrome [SIRS], sepsis or septic shock) and no other identifiable source of infection. The definition is based on the case definition of invasive bacterial disease from the Active Bacterial Core Surveillance, a collaboration between the Centers for Disease Control and Prevention and several state health departments and universities participating in the Emerging Infections Program network.[21,26]

Although IED affects all age categories, adults aged 60 years or older have an increased risk of developing IED, including bacteremia and sepsis, which can be either community-acquired, hospital-acquired or healthcare-associated. In a retrospective population-based study in the United States from 1998 to 2007, the incidence of *E. coli* bloodstream infections was shown to increase markedly with age from 60 years onwards. Overall incidences of *E. coli* bloodstream infections for adults aged ≥60 years were estimated at 136 to 152/100,000 person-years; by age stratum, incidences were estimated at 100/100,000 person-years for patients aged 60 to 79 years, and 300/100,000 person-years for patients aged ≥80 years. In this study, most cases were community-acquired (59.4%); the remainder were healthcare-associated (31.7%) or hospital-acquired (8.9%). The urinary tract was the most common primary source of infection (79.8%), followed by the gastrointestinal tract (8.7%), the respiratory tract (3.0%) and other sites

(1.3%).[11,12] In a retrospective study in England from April 2012 to March 2014, the most common underlying cause of bacteremia was infection of the genital/urinary tract (41.1%; 27,328/66,512), of which 98.4% (26,891/27,328) were urinary tract infections (UTIs).[13] Another US population-based cohort study showed that the incidence of community-acquired ExPEC bloodstream infections was 150/100,000 person-years in adults aged ≥65 years (1998-2001), and 452/100,000 person-years in adults aged ≥85 years.[17] According to data extracted from the Public Health England voluntary surveillance database for bacteremia caused by *E. coli* between 2010 and 2014 in England, Wales and Northern Ireland, the highest incidence rate of *E. coli* bacteremia was in patients aged ≥75 years (350.4/100,000 person-years), followed by patients aged 65 to 74 years (120.1/100,000 person-years).[6]

At present, there is no vaccine available to prevent IED. ExPEC10V is being developed to prevent IED in adults 60 years of age and older. The serotypes comprising the ExPEC10V vaccine (O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75) were selected to address invasive disease caused by the majority of clinically relevant ExPEC strains that also represent the majority of ExPEC isolates causing antimicrobial resistant IED, including ST131. The selected serotypes are generally the ten most prevalent ExPEC O-serotypes causing bloodstream infections in the older adult and elderly population and are responsible for approximately 70% of bloodstream infections caused by ExPEC.[7,8]

For the most comprehensive nonclinical and clinical information regarding ExPEC10V, refer to the latest version of the Investigator's Brochure for ExPEC10V.[3]

The term “study vaccination” or “study vaccine” throughout the protocol, refers to either ExPEC10V, ExPEC4V, Prevnar 13®, or placebo.

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

The term "participant" throughout the protocol refers to the common term "subject".

2.1. Study Rationale

The ExPEC10V vaccine is based on O-antigen PS structures that are part of the lipopolysaccharide located at the surface of the gram-negative bacterial outer membrane. These highly immunogenic surface structures induce O-antigen specific antibodies capable of inducing bacterial killing by opsonophagocytosis. The *E. coli* O-antigen PS structures included in the ExPEC10V vaccine are conjugated to the carrier protein, EPA. Based on prevalence and antibiotic resistance data collected in *E. coli* surveillance studies conducted in the United States and several European countries, the following ten *E. coli* O-serotypes, responsible for approximately 70% of bloodstream infections caused by ExPEC, were selected for inclusion in ExPEC10V: O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75.[7,8]

Since the mechanism of action of conjugate vaccines in the prevention of invasive disease is not expected to be affected by antibiotic resistance mechanisms, the sponsor believes that ExPEC10V

vaccine will provide protection against IED caused by drug-resistant and susceptible O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 serotypes.

ExPEC10V is being developed based on the sponsor's preceding clinical experience with ExPEC4V, an earlier vaccine candidate which comprised a subset of four of the *E. coli* O-antigen conjugates (O1A, O2, O6A and O25B) also found in ExPEC10V. The ExPEC4V vaccine has been evaluated in two completed Phase 1 clinical studies (GVXN-EC4V and 63871860BAC1001), one completed Phase 2 clinical study (63871860BAC2003) and one ongoing Phase 2 clinical study (63871860BAC2001; long-term follow-up [LTFU] completed and Clinical Study Report [CSR] in preparation). Based on the results from these three studies, ExPEC4V was well-tolerated by the study participants and no vaccine-related safety signals were observed at doses up to 16 µg PS per serotype (O1A, O2, O6A and O25B). Most adverse events (AEs) were Grade 1 and 2, very few Grade 3 AEs were reported. Late-onset solicited local AEs (AEs which start after Day 5 post-vaccination) were observed mainly with the higher doses of ExPEC4V. In each study, the ExPEC4V vaccine was shown to be immunogenic, demonstrating a dose-dependent vaccine immune response, and O-antigen specific immunoglobulin G (IgG) titer increases, as measured by enzyme-linked immunosorbent assay (ELISA). Functional activity of the antibodies was demonstrated with an ExPEC4V-optimized opsonophagocytic killing assay (OPA). Co-analysis of ELISA and OPA test results showed correlation between the assay responses (Pearson correlation coefficients ≥0.61 and ≥0.48 for Day 30 and Day 360, respectively in the Phase 2 clinical study [63871860BAC2001]), substantiating the use of ELISA as a primary measure of ExPEC4V antibody titers and to predict functional antibody activity. Analysis of the immunogenicity data from study 63871860BAC2001 has demonstrated the durability of the immune response through 3 years after vaccination with ExPEC4V.

ExPEC10V includes a total of ten serotypes and will increase coverage from 50% (ExPEC4V) to approximately 70% of bloodstream infections caused by ExPEC in adults aged 60 years and older.

The sponsor has also performed a systematic literature review and explored the Kaiser Permanente Northwest (KPNW) electronic health record database and the Clinical Practice Research Datalink (CPRD) to identify underlying conditions associated with an increased risk for IED. The data mining efforts identified a history of UTI as a key risk factor for developing IED.[29] Cohort 2 will include participants in stable health with a history of UTI in the past 5 years and will be included in the study to support the plan for late stage development of ExPEC10V.

2.2. Background

Nonclinical Studies

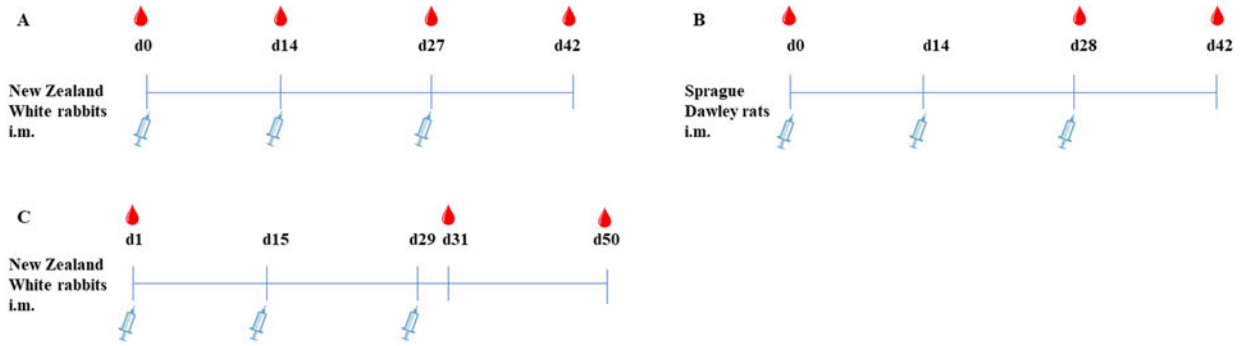
Immunogenicity Profile

The immunogenicity of the vaccine candidate ExPEC10V was evaluated in two preclinical models using New Zealand White (NZW) male and female rabbits (DS-TEC-130940, TOX13465) and Sprague-Dawley female rats (DS-TEC-130942).

For both species, the animals received three intramuscular (IM) vaccinations with ExPEC10V or saline control (0.9% [weight/volume] sodium chloride solution) 2 weeks apart (Figure 4, A, B and C).

The antibody levels induced by each of the O-serotypes included in the vaccine and the carrier protein EPA were measured by ELISA using specific O-lipopolysaccharide as coating material. The antibody titers were reported as half maximal effective concentration (EC_{50}) values based on duplicates of 12-step titration curves plotted in a 4-parameter logistic nonlinear regression model.

Figure 4: Vaccinations Schedule and In Vitro Read-Outs (DS-TEC-130940, DS-TEC-130942 and TOX13465)



(A) NZW female rabbits received three IM vaccinations administered 2 weeks apart (Days 0, 14 and 27). Antibody levels were determined by ELISA on Day 0 (pre-vaccination) and Days 14, 27 and 42 (post-vaccination).
(B) Sprague-Dawley female rats received three IM vaccinations administered 2 weeks apart (Days 0, 14 and 28). Antibody levels were determined by ELISA on Day 0 (pre-vaccination) and Days 28 and 42 (post-vaccination).
(C) NZW male and female rabbits received three IM vaccinations administered two weeks apart (Days 1, 15, and 29). Antibody levels were determined by ELISA on Day 1 (pre-vaccination) and Days 31 and 50 (post-vaccination).

Table 3: Experimental Groups Description in New Zealand White Female Rabbits (DS-TEC-130940)

Experimental Groups	Dosing (µg PS) O1A:O2:O4:O6A:O8:O15:O16:O18A:O25B:O75	Number of Animals
Group 1 (high dose)	8:8:8:8:8:8:8:8:16:8	7
Group 2 (medium dose)	8:4:4:8:4:4:4:4:16:4	7
Group 3 (low dose)	0.8:0.4:0.4:0.8:0.4:0.4:0.4:0.4:1.6:0.4	7
Group 4 (control)	0.9% (w/v) sodium chloride solution	7

w/v weight by volume

In DS-TEC-130940 Study, the three doses of ExPEC10V (high, medium and low, see Table 3), administered via IM injection on Days 0, 14, and 27 were immunogenic in female rabbits. Significantly higher IgG antibody titers were observed in the vaccinated animals when compared to the group that received only saline for all O-serotypes included in the vaccine, except for O15. For the O15 conjugate, high levels of antibodies were induced by the three doses of ExPEC10V on Days 14, 27 and 42, however, due to high titers observed for one of the animals from the control group, the differences did not reach statistical significance.

In addition, dose comparisons show that the high and medium doses of ExPEC10V induced higher antibody responses when compared to the low dose already on Day 14 post-vaccination (ie, 2

weeks after prime) for most of the conjugates. In contrast, for O1A, O2, O4, O16, O25B and O75 conjugates, the low dose of the vaccine induced a better boost effect on Day 42 post-vaccination (ie, after the third dose of the vaccine) in rabbits.

Table 4: Experimental Groups Description in Sprague-Dawley Female Rats (DS-TEC-130942)

Experimental Groups	Dosing (µg PS) O1A:O2:O4:O6A:O8:O15:O16:O18A:O25B:O75	Number of Animals
Group 1 (high dose)	8:4:4:8:4:4:4:4:16:4	10
Group 2 (medium dose)	0.8:0.4:0.4:0.8:0.4:0.4:0.4:0.4:1.6:0.4	10
Group 3 (low dose)	0.08:0.04:0.04:0.08:0.04:0.04:0.04:0.04:0.16:0.04	10
Group 4 (control)	0.9% (w/v) sodium chloride solution	10

w/v weight by volume

In DS-TEC-130942 Study, the three doses of ExPEC10V (high, medium and low, see Table 4), were immunogenic in female rats when administered IM on Days 0, 14 and 28; significantly higher IgG antibody titers were observed for all conjugates included in the vaccine when compared to saline control at least at one of the time points investigated and vaccine doses tested, except for O16. For O16 conjugate, low antibody responses were observed with all three doses of ExPEC10V tested.

In addition, the high dose of ExPEC10V induced significantly higher antibody responses after the prime and first boost dose (Day 28 post-vaccination) when compared to the medium and low dose of the vaccine for most of the conjugates (O1A, O2, O4, O6A, O15, O18A and O25B). For O8, O16 and O75 conjugates, similar levels of antibodies were induced with the three different doses of ExPEC10V.

Table 5: Experimental Groups Description in New Zealand White Male and Female Rabbits (TOX13465)

Experimental Group	Treatment	Dose Level	Dosing Days	Number of Animals			
				Main Day 31 Kill		Recovery Day 50/51 Kill	
				Males	Females	Males	Females
Group 1	Control	0	1, 15, 29	5	5	5	5
Group 2	ExPEC10V	105.6 µg total PS[a]	1, 15, 29	5	5	5	5

Control=0.9% (weight by volume [w/v]) sodium chloride solution, sterile for injection.
[a] Each dose (0.6 mL dosing volume) contained 9.6:9.6:9.6:9.6:9.6:9.6:9.6:9.6:19.2:9.6 µg PS for serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B, O75, respectively (176 µg total PS/mL). Each dose contained 382.8 µg EPA protein (638 µg EPA/mL).

The immunogenicity of ExPEC10V vaccine was evaluated in NZW rabbits as part of a toxicology study (TOX13465). In this study, NZW rabbits (males and females) received three IM vaccinations of the ExPEC10V vaccine or saline control administered two weeks apart (Days 1, 15, and 29, see Table 5). ExPEC10V was confirmed to be immunogenic in rabbits; significantly higher antibody responses were observed against all O-conjugates and the carrier protein EPA on Day 31 (ie, two days after the third vaccination) and Day 50 (ie, 21 days after the third vaccination) when compared to saline control.

Toxicology

A single-dose pilot toxicity and local tolerance study (Study TOX13466, non-GLP) with ExPEC10V was conducted in female NZW rabbits. One group (n 2) received an IM injection (on Day 0) of the control (saline), and a second group (n 4) received an IM injection of ExPEC10V at 105.6 µg total PS/dose[a] using a dosing volume of 0.6 mL (176 µg PS/mL). Necropsy was performed on Day 2.

There were no mortalities observed. In addition, there were no vaccine-related effects noted for clinical observations (including injection site effects using Draize scoring), body weight, food consumption, and body temperature. Histopathologically, there were no vaccine-related changes observed at the administration site or draining (iliac) lymph node. A minimal increase in germinal center formation in the spleen was observed in one out of four treated animals (Day 2), and was considered a normal, immunological response to the injected vaccine. Overall, the administration of a single IM dose of ExPEC10V to female rabbits was well-tolerated.

To support the proposed Phase 1/2a clinical study, a 4-week GLP intermittent IM repeat-dose toxicity study in rabbits (Study TOX13465) was conducted to evaluate the nonclinical safety profile of ExPEC10V in NZW rabbits. The main objectives of this toxicology study were:

- To assess potential toxicity and local tolerance of ExPEC10V when administered to NZW rabbits by IM injection once every 2 weeks for 4 weeks (ie, three injections).
- To evaluate the reversibility, persistence or delayed occurrence of any adverse effects of the vaccine regimen during a 3-week treatment-free period following the last injection.

ExPEC10V was administered once every two weeks at 105.6 µg total PS/dose (with a total of three IM injections; for details, see Table 5). ExPEC10V was shown to be immunogenic in the animals. A transient acute phase inflammatory response was observed, characterized by higher plasma proteins including C-reactive protein (CRP), and microscopic findings at the injection sites (inflammation), draining iliac lymph node, and spleen (increased lymphoid cellularity of the germinal centers). After a 3-week recovery period, plasma proteins had returned to baseline, and the microscopic findings at the injection sites, draining lymph node, and spleen showed ongoing recovery. All vaccine-related effects noted were considered to reflect a normal immune/inflammatory response to the administered vaccine and were not considered to be adverse.

Clinical Studies

This will be the first administration of ExPEC10V in humans; therefore, no previous clinical experience is available. However, ExPEC10V is being developed based on the sponsor's preceding clinical experience with ExPEC4V, an earlier vaccine candidate which has been evaluated in two completed Phase 1 clinical studies (GVXN-EC4V and 63871860BAC1001) and two Phase 2

[a] 9.6: 9.6: 9.6: 9.6: 9.6: 9.6: 9.6: 9.6: 19.2: 9.6 µg polysaccharide (PS) per dose (0.6 mL injection volume), for respectively O-serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75.

clinical studies (the ongoing study 63871860BAC2001 [LTFU completed and CSR in preparation] and the completed study 63871860BAC2003). Refer to Section 2.2.1, Comparator Vaccines for more details.

2.2.1. Comparator Vaccines

ExPEC4V (JNJ-63871860)

ExPEC10V is being developed based on the sponsor's preceding clinical experience with ExPEC4V, an earlier vaccine candidate which comprised a subset of four of the O-antigen conjugates (O1A, O2, O6A and O25B) also found in ExPEC10V. A summary of the studies conducted for ExPEC4V is provided in Table 6.

Table 6: Clinical Studies Conducted Under ExPEC4V Development

Study *Phase*	Study Population *Country(s)*	Study Design	Number of Participants		Endpoints	Status
			ExPEC4V (All Doses)	Placebo		
GVXN-EC4V FIH *Phase 1*	Women ≥18 to ≤70 years *Country: Switzerland*	Randomized, single blind, placebo controlled multicenter study	99	95	Safety, immunogenicity, and efficacy in prevention of UTI	Completed
63871860BAC1001 *Phase 1*	Healthy Japanese men and women ≥20 years *Country: Japan*	Randomized, double blind, placebo controlled parallel group single center study	36	12	Safety and immunogenicity	Completed
63871860BAC2001 Dose finding *Phase 2*	Men and women in stable health ≥18 years *Country: US*	Randomized, double blind, placebo controlled multicenter study	757	86	Safety and immunogenicity	LTFU completed and CSR in preparation
63871860BAC2003 Safety/ Immunogenicity study with new ExPEC4V CTM *Phase 2*	Healthy men and women ≥18 years *Country: US*	Randomized, double blind, placebo controlled study	75	25	Safety and immunogenicity of Phase 3 CTM	Completed

CSR Clinical Study Report; CTM clinical trial material; FIH first in human; LTFU long term follow up; US United States; UTI urinary tract infection

Based on the results from these studies, ExPEC4V was well-tolerated by the study participants and no vaccine-related safety signals were observed at doses up to 16 µg PS per serotype (O1A, O2, O6A and O25B). ExPEC4V vaccine was shown to be immunogenic, demonstrating a dose-dependent vaccine immune response. Analysis of the durability of the vaccine-induced immune responses from study 63871860BAC2001 showed that antibody responses were maintained for 3 years post-vaccination with ExPEC4V.

For further information regarding ExPEC4V, refer to the Investigator Brochure.[2]

Prevnar 13®

Prevnar 13 is an approved vaccine indicated for the active immunization for the prevention of pneumonia and invasive disease caused by *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F and 23F. The safety and reactogenicity profile of Prevnar 13 has been shown to be acceptable in adult and elderly studies. For more information regarding Prevnar 13, refer to the package insert.[9]

2.3. Benefit/Risk Assessment

2.3.1. Known Benefits

The clinical benefits of ExPEC10V and ExPEC4V are not yet established.

Currently, there are no effective vaccines for IED and no efficacy can be concluded from current data. The overall benefit and risk balance for individual participants thus cannot be ascertained. Participants must be informed that this vaccine has not yet been tested to determine whether it is effective, and it should be assumed that it is not the case until clinical studies are conducted to demonstrate its effectiveness.

In Cohort 1, some participants (Group 5; see Table 1) will receive Prevnar 13 as study vaccination on Day 1 and the other participants (Groups 1-4; see Table 1) will be offered Prevnar 13 vaccination outside of the study after final analysis database lock (Day 181). Prevnar 13 is an approved and marketed vaccine indicated for the active immunization for the prevention of pneumonia and invasive disease caused by *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F and 23F. The safety and reactogenicity profile of Prevnar 13 has been shown to be acceptable in adult and elderly studies.[9]

2.3.2. Potential Benefits

The individual benefit from ExPEC10V and ExPEC4V vaccination for the participants at the current development stage is not known. Although participants may benefit from clinical testing and physical examination, they may receive no direct benefit from participation. Others may benefit from the knowledge gained in this study that may aid in the development of ExPEC10V, which will provide protection against IED.

2.3.3. Known Risks

There are no known risks related to ExPEC10V.

The available nonclinical and safety data with ExPEC4V, a predecessor vaccine, support the conclusion that the ExPEC4V vaccine has an acceptable safety profile with no emerging significant safety concerns to date.

For risks related to Prevnar 13, please refer to the package insert.[9]

2.3.4. Potential Risks

The following potential risks for ExPEC10V and ExPEC4V will be monitored during the study and are specified in the protocol:

Risks Related to Vaccination

In general, IM injection may cause local itching, warmth, pain, tenderness, erythema, swelling, arm discomfort or bruising of the skin at vaccine injection sites. Participants may exhibit general signs and symptoms associated with administration of a vaccine, or vaccination with active controls, including fever, chills, rash, myalgia, nausea/vomiting, headache, dizziness, arthralgia, general itching, and fatigue. These side effects are generally short-term and do not require treatment.

Syncope can occur in association with administration of injectable vaccines. Syncope can be accompanied by falls. Procedures should be in place to avoid falling injury. If syncope develops, participants should be observed until the symptoms resolve. Fear of injection might lead to fainting and fast breathing.

Participants may have an allergic reaction to the vaccination. An allergic reaction may cause a rash, urticaria or even anaphylaxis. Severe reactions are rare. Potential participants with a known allergy, or history of anaphylaxis or other serious adverse reactions to vaccines or their excipients (including any of the constituents of the study vaccine) will be excluded from the study.

Pregnancy and Birth Control

The effect of this vaccine on a fetus or nursing baby is unknown. This study will enroll postmenopausal women (defined as no menses for 12 months without an alternative medical cause) or women who are not intending to conceive by any methods.

Risks from Blood Draws

As with all clinical studies requiring blood sampling, there are risks associated with venipuncture and multiple blood sample collection. Blood drawing may cause pain, tenderness, bruising, bleeding, dizziness, vaso-vagal response, syncope, and, rarely, infection at the site where the blood is taken. The total blood volume to be collected is considered to be an acceptable amount of blood over this time period for the population in this study (see Section 4.2.3, Study-Specific Ethical Design Considerations).

Unknown Risks

There may be other risks that are not known. If any significant new risks are identified, the principal investigator (PI) and participants will be informed.

2.3.5. Overall Benefit/Risk Assessment

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

- Only participants who meet all inclusion criteria and none of the exclusion criteria (specified in Section 5) will be allowed to participate in this study. The selection criteria include adequate provisions to minimize the risk and protect the well-being of participants in the study.
- Safety will be closely monitored throughout the study:

  In general, safety evaluations will be performed at scheduled visits during the study, as indicated in Section 1.3 Schedule of Activities.

  After the vaccination, participants will remain at the study site for a certain period and will be closely observed by study staff. Necessary emergency equipment and medications must be available in the clinic to treat severe allergic reactions. Participants will use an electronic diary (ediary) to document solicited local and systemic symptoms. Details are provided in Section 8.2, Adverse Events and Serious Adverse Events.

  In Phase 1 of Cohort 1, initial four sentinel participants at each dose level will undergo safety follow-up by study staff 24 hours after the vaccination by telephone and all participants will undergo safety follow-up by study staff 48 hours after the vaccination by telephone, as indicated in Section 1.3 Schedule of Activities.

  The investigator or the designee will document unsolicited AEs as indicated in Section 8.2, Adverse Events and Serious Adverse Events and Section 10.3 Appendix 3 Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting.

  Any clinically significant abnormalities (including those persisting at the end of the study/early withdrawal) will be followed by the investigator until resolution or until a clinically stable endpoint is reached.
- Several safety measures are included in this protocol to minimize the potential risk to participants, including the following:

  In Cohort 1, participants will be enrolled in a staggered approach following stepwise dose-escalating procedures. Initially four sentinel participants will be vaccinated at each dose level followed by a 24-hour safety review. If deemed safe, an additional 24 participants will be randomized at each dose level. After dosing of 28 participants at each dose level, a Data Review Committee (DRC) review will be performed to evaluate the 14-day post-vaccination safety data before progressing to the next dose level or Phase 2a. For more details refer to Section 4.1 Overall Design.

  For all participants, there are pre-specified rules that would result in pausing of further vaccinations if predefined conditions occur (see Section 9.7 Study Vaccination Pausing Rules for more details), preventing exposure of new participants to study vaccination until the DRC reviews all safety data (see Committee Structure in Section 10.2 Appendix 2, Regulatory, Ethical, and Study Oversight Considerations).

Contraindications to vaccination are included in Section 7.1.1 Contraindications to Study Vaccination.

More detailed information about the known and expected benefits and risks of ExPEC10V may be found in the ExPEC10V Investigator's Brochure.[3]

3. OBJECTIVES AND ENDPOINTS

This study's aim is to assess the safety, reactogenicity, and immunogenicity of 3 different doses of ExPEC10V and to select the optimal dose for further clinical development (Cohort 1). Cohort 2 is aimed to expand the dataset supporting the short- and long-term safety and immunogenicity of the optimal dose of ExPEC10V, selected from the primary analysis results of Cohort 1. Cohort 2 will include approximately 420 male and female participants in stable health with a history of urinary tract infection (UTI) in the past 5 years and will be included in the study to support the plan for late stage development of ExPEC10V.

3.1. Cohort 1 - Phase 1/2a Observer-Blind Period With Open-Label Long-Term Follow-up Period (N=404)

Objectives	Endpoints
Primary	
• To evaluate the safety and reactogenicity of different doses of ExPEC10V in participants ≥60 to ≤85 years of age	• Solicited local and systemic AEs collected for 14 days post-vaccination (from Day 1 to Day 15) • Unsolicited AEs collected from the administration of the study vaccine until 29 days post-vaccination (from Day 1 to Day 30) • Serious adverse events (SAEs) collected from the administration of the study vaccine until Day 181
• To evaluate the dose-dependent immunogenicity of ExPEC10V on Day 15 in participants ≥60 to ≤85 years of age	• Antibody titers for ExPEC10V, as determined by multiplex electrochemiluminescent (ECL)-based immunoassay and multiplex opsonophagocytic assay (MOPA) on Day 15

Objectives	Endpoints
Secondary	
• To evaluate the correlation between multiplex ECL-based immunoassay (total antibody) and MOPA (functional antibody) serum titers on Day 15	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 15
• To evaluate the dose-dependent immunogenicity of ExPEC10V on Days 30 and 181 in participants ≥60 to ≤85 years of age	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Days 30 and 181
• To evaluate, in the LTFU period, the safety of the ExPEC10V dose selected for further clinical development based on the Day 30 primary analysis in participants ≥60 to ≤85 years of age	• SAEs related to the study vaccine or study procedures collected from Day 182 until the end of the study
• To evaluate, in the LTFU period, the immunogenicity of the ExPEC10V dose selected for further clinical development based on the Day 30 primary analysis	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826)

3.2. Cohort 2 - Double-Blind Period With Double-Blind Long-Term Follow-up Period (N=±420)

Objectives	Endpoints
Primary	
• To evaluate the safety and reactogenicity of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• Solicited local and systemic AEs collected for 14 days post-vaccination (from Day 1 to Day 15) • Unsolicited AEs collected from the administration of the study vaccine until 29 days post-vaccination (from Day 1 to Day 30) • SAEs collected from the administration of the study vaccine until Day 181
• To evaluate the immunogenicity of the selected dose of ExPEC10V on Day 30 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 30

Objectives	Endpoints
Secondary	
• To evaluate the correlation between multiplex ECL-based immunoassay (total antibody) and MOPA (functional antibody) serum titers on Day 30 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA on Day 30
• To evaluate the immunogenicity of the selected dose of ExPEC10V on Days 15 and 181 in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay on Days 15 and 181 and MOPA on Day 181
• To evaluate, in the LTFU period, the safety of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• SAEs related to the study vaccine or study procedures collected from Day 182 until the end of the study
• To evaluate, in the LTFU period, the immunogenicity of the selected dose of ExPEC10V in participants ≥60 years of age with a history of UTI in the past 5 years	• Antibody titers for ExPEC10V, as determined by multiplex ECL-based immunoassay and MOPA at Year 1 (Day 366)
Exploratory	
• To evaluate the effect of ExPEC10V on the intestinal (stool) microbiome by metagenomic analyses	• Metagenomics of stool samples from a selected subset[a] of participants to evaluate the effect of ExPEC10V on: Prevalence of pathogens (eg, *Clostridium difficile*) in the intestinal flora Prevalence of ExPEC10V serotypes in the intestinal flora

Refer to Section 8, Study Assessments and Procedures for evaluations related to endpoints.

HYPOTHESIS

No formal hypothesis testing will be done. One of the three ExPEC10V doses tested in Cohort 1 will be selected for further clinical development based on the safety (through Day 30) and immunogenicity data (Day 15 results obtained by multiplex ECL-based immunoassay and MOPA). The dose selection will consider the totality of the evidence available at the time of the primary analysis (Day 30) for Cohort 1, including an immunogenicity dose-selection algorithm that will guide the decision.

[a] A subset of 33% of participants selected at randomization on Day 1 (using the interactive web response system [IWRS]).

In Cohort 2, the safety and immunogenicity of the ExPEC10V dose selected from Cohort 1 will be further evaluated in participants in stable health with a history of UTI in the past 5 years.

4. STUDY DESIGN

4.1. Overall Design

This is a Phase 1/2a randomized, multicenter, interventional study including two cohorts.

For Cohort 1, the study will have an observer-blind, active-controlled design, and a total of 404 adult participants aged ≥60 to ≤85 years in stable health with or without a history of UTI will be included.

For Cohort 2, the study will have a double-blind, placebo-controlled design, and a total of approximately 420 adult participants aged ≥60 years in stable health with a history of UTI in the past 5 years will be included.

A DRC will be commissioned for this study. Refer to Committees Structure in Section 10.2, Appendix 2: Regulatory, Ethical, and Study Oversight Considerations for details. The study vaccination pausing rules are provided in Section 9.7.

Refer to Section 10.6 for guidance on study conduct during the Coronavirus Disease 2019 (COVID-19) pandemic.

4.1.1. Cohort 1

The study design for Cohort 1 is comprised of three periods: a maximum 28-day screening period, an observer-blind 181-day follow-up period with vaccination on Day 1 and an open-label LTFU period which will last from Day 182 until 5 years (Day 1826) post-vaccination (Figure 1). Only participants from the ExPEC10V selected dose group (approximately 100 participants) and participants from the Prevnar 13 group (approximately 52 participants) will progress to the LTFU period. All participants randomized to either of the three doses of ExPEC10V as well as ExPEC4V participants (Groups 1-4; see Table 1) will be offered Prevnar 13 vaccination outside of the study after the final analysis database lock (Day 181) in order to follow Advisory Committee for Immunization Practices (ACIP) recommendations published in September 2014.[1]

During the open-label LTFU period, participation in another investigational study is allowed only with prior approval of the sponsor. If participating in another investigational study, it is recommended to administer the investigational product 45 days before the next planned study visit.

The end of Cohort 1 will be the last participant's Year 5 visit (Day 1826).

Phase 1

In Phase 1 of Cohort 1, 84 participants will be enrolled in a staggered approach following stepwise dose-escalating procedures with safety evaluations in place before progressing from one step to the next. An internal DRC will be commissioned for this study to review the physical examination

data (baseline as well as targeted), baseline demographic data and the 14-day post-vaccination safety data (including solicited local and systemic AEs, unsolicited AEs, SAEs, clinical laboratory data and vital signs) of these 84 Phase 1 participants. If deemed necessary for safety review, the DRC may request randomization codes and review unblinded data.

In this phase of the study, participants will be enrolled and randomized in six steps:

- Step 1: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V low dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups. These four sentinel participants will receive a single IM injection of the ExPEC10V low dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.
- Step 2: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V low dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups. These 24 participants will receive a single IM injection of the ExPEC10V low dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.
- Step 3: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V medium dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups. These four sentinel participants will receive a single IM injection of the ExPEC10V medium dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.
- Step 4: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V medium dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups. These 24 participants will receive a single IM injection of the ExPEC10V medium dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.
- Step 5: Four sentinel participants will be enrolled and randomized; two participants in the ExPEC10V high dose group (Table 1), and one participant each in the ExPEC4V and Prevnar 13 groups. These four sentinel participants will receive a single IM injection of the ExPEC10V high dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.
- Step 6: Twenty-four participants will be enrolled and randomized; 18 participants in the ExPEC10V high dose group (Table 1), and three participants each in the ExPEC4V and Prevnar 13 groups. These 24 participants will receive a single IM injection of the ExPEC10V high dose, ExPEC4V or Prevnar 13 (Table 7) on Day 1.

The four sentinel participants at each of Steps 1, 3 and 5 will be contacted by telephone 24 hours post-vaccination to collect safety information. The blinded 24-hour post-vaccination safety data in each group of four sentinel participants will be reviewed by the PI, study responsible physician (SRP) and sponsor medical lead (SML). Randomization of additional participants for the next step will be halted until this Day-2 sentinel safety evaluation is completed (Figure 3).

In the absence of any clinically significant findings, upon decision by the PI, SRP and SML, an additional 24 participants (for Steps 2, 4 and 6) will be enrolled and randomized in parallel in a ratio of 6:1:1 to one of three study vaccination groups (Table 1, Figure 3) to receive a single IM injection of ExPEC10V (low dose in Step 2, medium dose in Step 4 and high dose in Step 6; Table 7), ExPEC4V or Prevnar 13 on Day 1.

After vaccination of an additional 24 participants at each dose level (low dose in Step 2, medium dose in Step 4, and high dose in Step 6), 14-day post-vaccination safety data of all 28 (4+24) participants will be reviewed by the DRC before progressing to the next dose level or Phase 2a. Further randomization will be halted until the DRC safety evaluation is completed at each step (Figure 3).

Phase 2a

Based on acceptable safety and reactogenicity (in the absence of any safety concerns or any events meeting a specific study vaccination pausing rule [Section 9.7]) as determined by the DRC after the review of 14-day post-vaccination safety data for the initial 84 participants, the remaining 320 participants from Cohort 1 will be randomized and dosed in Phase 2a. These additional 320 participants will be enrolled and randomized in parallel in a ratio of 2:2:2:1:1 to one of the five study vaccination groups to receive a single IM injection of either ExPEC10V (1 of 3 doses), ExPEC4V or Prevnar 13 on Day 1 (Table 1).

In addition to performing the 14-day safety review for the initial 84 participants, the DRC will also evaluate safety data of Cohort 1 over the course of the study and review any events that meet a specific study vaccination pausing rule (Section 9.7) or any other safety issue that may arise. The DRC will review blinded data first but may also review unblinded data, if deemed necessary.

The overall study design diagram for Cohort 1 (Phase 1 and Phase 2a) is provided in Section 1.2, Schema.

Key safety assessments include solicited local and systemic AEs, unsolicited AEs, SAEs, physical examinations, vital sign measurements, and clinical laboratory tests (refer to Section 8.1, Safety Assessments and Section 8.2, Adverse Events and Serious Adverse Events). Key immunogenicity assessments of collected sera will include the assessment of ExPEC10V and ExPEC4V serotype-specific total IgG antibody levels elicited by the vaccine as measured by multiplex ECL-based immunoassay, and ExPEC10V and ExPEC4V serotype-specific functional antibodies as measured by MOPA. Immunogenicity assessments of pneumococcal antibody titers elicited by Prevnar 13 will not be performed (refer to Section 8.4, Immunogenicity Assessments).

For Cohort 1, the primary analysis will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. The final analysis will occur when all participants have completed the Day 181 visit or have discontinued earlier. The study sites will be informed after the final analysis database lock which participants should progress to the LTFU period (ExPEC10V selected dose group and Prevnar 13 group) and for which participants the Day 181 visit will be the last on-site visit. For all participants, a telephone contact will be scheduled to inform them whether they will be progressing to the LTFU period. For participants not progressing to the LTFU period, that telephone contact will be their last study visit.

For participants progressing to the open-label LTFU period, yearly follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to

the time of the visit at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826) after vaccination.

4.1.2. Cohort 2

After completion of the Phase 1/2a primary analysis and ExPEC10V dose selection from Cohort 1, the safety, reactogenicity, and immunogenicity of the selected dose of ExPEC10V (based on the primary analysis results of Cohort 1) will be evaluated in Cohort 2, including participants aged ≥60 years in stable health with a history of UTI in the past 5 years. For Cohort 2, the study will have a double-blind, placebo-controlled design, and a total of approximately 420 participants will be enrolled and randomized in parallel in a 2:1 ratio (280 participants in the ExPEC10V group and 140 in the placebo group).

The study design for Cohort 2 is comprised of three periods: a maximum 28-day screening period, a double-blind 181-day follow-up period with vaccination on Day 1, and a double-blind LTFU period which will last from Day 182 until 1 year (Day 366) post-vaccination (Figure 2). All participants in Cohort 2 will progress to the LTFU period.

All participants will receive a single IM injection of either the selected dose of ExPEC10V or placebo on Day 1 per the assigned study vaccination groups (Table 2). If a specific study vaccination pausing rule (Section 9.7) has been met or if any other safety issue may arise, the DRC will be convened to evaluate safety data of Cohort 2. The DRC will review blinded data first but may also review unblinded data, if deemed necessary.

During the double-blind LTFU period, participation in another investigational study is allowed only with prior approval of the sponsor. If participating in another investigational study, it is recommended to administer the investigational product 45 days before the next planned study visit.

The end of Cohort 2 will be the last participant's Year 1 visit (Day 366) in Cohort 2.

The end of study will be the last participant's Year 5 visit (Day 1826) in Cohort 1.

The overall study design diagram for Cohort 2 is provided in Section 1.2, Schema.

Key safety assessments include solicited local and systemic AEs, unsolicited AEs, SAEs, physical examinations, and vital sign measurements (refer to Section 8.1, Safety Assessments and Section 8.2, Adverse Events and Serious Adverse Events). Key immunogenicity assessments of collected sera will include the assessment of ExPEC10V serotype-specific total IgG antibody levels elicited by the vaccine as measured by multiplex ECL-based immunoassay and ExPEC10V serotype-specific functional antibodies as measured by MOPA (refer to Section 8.4, Immunogenicity Assessments).

In addition, a stool sample analysis will be performed in a selected subset of participants to evaluate the effect of ExPEC10V on the prevalence of pathogens (eg, *Clostridium difficile*) and

ExPEC10V serotypes in the intestinal flora using metagenomics (refer to Section 8.5, Other Evaluations).

For Cohort 2, the primary analysis will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. The final analysis will occur when all participants have completed the Day 181 visit or have discontinued earlier. For all participants, follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to the time of the visit at Year 1 (Day 366) after vaccination (refer to Section 9.5, Planned Analyses).

4.2. Scientific Rationale for Study Design

4.2.1. Cohort 1

Observer-Blind

An observer-blind design is needed when the physical appearance of the candidate vaccine and control vaccine differs. In this study, due to the differences in the appearance of the study vaccine used, ExPEC10V and ExPEC4V in comparison to Prevnar 13, an observer-blind study design will be used. There will be a designated unblinded pharmacist(s) and/or a qualified staff member(s) at the site with primary responsibility for study vaccine preparation and administration; they will not participate in any other study evaluation. Separate blinded site staff or qualified professional staff, including the investigator(s), will perform the observation on the participants and the participants will also be blinded.

Active Controls

Two of the five study vaccination groups will consist of active controls:

- One group will use ExPEC4V (Table 7, dose of 4:4:4:8 µg PS based on the primary analysis results [Day 30] from the 63871860BAC2001 study) to guide decision making in case any unexpected immunogenicity and/or safety results are observed in the ExPEC10V groups with focus on the four serotypes (O1A, O2, O6A and O25B) tested in the ExPEC4V group.
- Another group will use Prevnar 13 as a reference group for safety and reactogenicity. Prevnar 13 is an approved vaccine indicated for the active immunization for the prevention of pneumonia and invasive disease caused by *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F and 23F. The safety and reactogenicity profile of Prevnar 13 has been shown to be acceptable in adult and elderly studies.[9] Participants previously vaccinated with Prevnar 13 (or other pneumococcal vaccines [PCVs]) or planning to receive any PCV before the final analysis (Day 181) will be excluded from Cohort 1.

Randomization

Randomization will be used to minimize bias in the assignment of participants to study vaccination groups, to increase the likelihood that known and unknown participant attributes (eg, demographic and baseline characteristics) are evenly balanced across study vaccination groups, and to enhance the validity of statistical comparisons across study vaccination groups.

4.2.2. Cohort 2

Double-Blind

Blinded study vaccination will be used to reduce potential bias during data collection and evaluation of safety and immunogenicity endpoints. There will be a designated unblinded pharmacist(s) and/or a qualified staff member(s) at the site with primary responsibility for study vaccine preparation; they will not participate in any other study evaluation. The vaccine administrator will be blinded and can perform other study evaluations.

Placebo Control

A placebo control will be used in Cohort 2 of the study to establish the frequency and magnitude of changes in safety and immunogenicity endpoints that may occur in the absence of active vaccination.

Prevnar 13 is being used as a reference group in Cohort 1 to guide the ExPEC10V dose selection in Phase 1, by comparing the safety and reactogenicity of ExPEC10V with that of Prevnar 13, an approved vaccine for the prevention of pneumonia and invasive disease caused by *Streptococcus pneumoniae* in older adults. In Cohort 2, after selection of the ExPEC10V dose from Cohort 1, a placebo control (saline solution) will be used to better characterize the safety and reactogenicity of ExPEC10V.

ExPEC4V is not included in Cohort 2 as this predecessor vaccine was included in Cohort 1 to guide decision for the dose selection of ExPEC10V in terms of immunogenicity and safety, based on the ExPEC4V data collected from previous studies.

Randomization

Randomization will be used to minimize bias in the assignment of participants to study vaccination groups, to increase the likelihood that known and unknown participant attributes (eg, demographic and baseline characteristics) are evenly balanced across study vaccination groups, and to enhance the validity of statistical comparisons across study vaccination groups.

4.2.3. Study-Specific Ethical Design Considerations

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

The primary ethical concern is the total blood volume to be collected from each participant which will not exceed the US Department of Health and Human Services (HHS) Office for Human Research Protections (OHRP), and US Food and Drug Administration (FDA) guidelines of 550 mL in any 8-week period.[5,10]

4.3. Justification for Dose

Selection of ExPEC10V Dose Groups

In Cohort 1 of the current study (VAC52416BAC1001), one single vaccination (0.5 mL) of the three different concentrations of ExPEC10V (O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75) will be evaluated; refer to the table below for more details.

Table 7: ExPEC Study Vaccination

Study Vaccination Group	O1A (µg)	O2 (µg)	O4 (µg)	O6A (µg)	O8 (µg)	O15 (µg)	O16 (µg)	O18A (µg)	O25B (µg)	O75 (µg)	EPA (µg)	PS (Total) (µg)
Low dose ExPEC10V	4	4	4	4	4	4	4	4	8	4	159	44
Medium dose ExPEC10V	8	4	4	8	4	4	4	4	16	4	217	60
High dose ExPEC10V	8	8	8	8	8	8	8	8	16	8	319	88
ExPEC4V	4	4	-	4	-	-	-	-	8	-	72	20

EPA a genetically detoxified form of exotoxin A derived from *Pseudomonas aeruginosa*; PS polysaccharide
ExPEC4V consists of the O antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O6A, and O25B separately bioconjugated to the EPA carrier protein.
ExPEC10V consists of the O antigen polysaccharides (PSs) of the ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the EPA carrier protein.
The EPA (µg) is calculated using a ratio of 0.276 for PS/EPA. However, the final EPA dose will be confirmed at the release.

The low, medium and high doses selected for evaluation of the 10-valent ExPEC10V vaccine (Table 7) are based on the immunogenicity (ELISA and OPA) and safety results of a previous ExPEC4V (4-valent) Phase 2 study (63871860BAC2001), where 4:4:4:8 and 8:8:8:16 µg PS doses (per serotype doses for O1A, O2, O6A and O25B, respectively) gave a robust immune response and reached a maximum or near-maximum immune response for all four serotypes on Day 15. One-year durability of the ExPEC4V antibody response, although demonstrated for both dose groups, was slightly improved for serotypes O1A and O6A in the 8:8:8:16 dose group. Thus, by extrapolation of the 4:4:4:8 and 8:8:8:16 ExPEC4V doses, the dose groups for the ten vaccine serotypes in the ExPEC10V vaccine are as follows (Table 7):

- the low dose group is formulated with 4 µg PS for nine serotypes (O1A, O2, O4, O6A, O8, O15, O16, O18A and O75), with serotype O25B at 8 µg,
- the high dose group has 8 µg PS for these nine serotypes and 16 µg for O25B, and
- the medium dose group was designed to maximize titers and durability of the dose response for serotypes O1A, O6A and O25B (8 µg, 8 µg and 16 µg, respectively), while maintaining the low dose of 4 µg for the remaining seven serotypes to minimize potential risks for immune interference and minimizing reactogenicity.

An acceptable safety profile using similar or higher PS/EPA doses has been demonstrated in clinical studies. The maximum amount of antigen evaluated in a study performed with a multivalent *E. coli* vaccine consisted of 300 µg PS and 400 µg EPA in 88 vaccinees.[14] The highest doses for PS and EPA to be tested in this study are 88 µg and 319 µg respectively

4.4. End of Study Definition

4.4.1. Cohort 1

4.4.1.1. Observer-Blind Period

A participant in Cohort 1 will be considered to have completed the observer-blind period of the study if he or she has completed Day 181. Participants who prematurely discontinue from the study for any reason before completion of the observer-blind period (Day 181) will not be considered to have completed the observer-blind period.

4.4.1.2. Long-Term Follow-Up Period

For participants in Cohort 1 progressing to the open-label LTFU period of the study, a participant will be considered to have completed the study if he or she has completed the open-label LTFU period (Year 5 visit, Day 1826).

The end of Cohort 1 is considered as the Year 5 visit (Day 1826) for the last participant in Cohort 1. The final data from the study site will be sent to the sponsor (or designee) after completion of the final participant visit at that study site, in the time frame specified in the clinical trial agreement.

4.4.2. Cohort 2

4.4.2.1. Double-Blind Period Until Day 181

A participant in Cohort 2 will be considered to have completed the double-blind period until Day 181 of the study if he or she has completed Day 181 of the study. Participants who prematurely discontinue from the study for any reason before Day 181 will not be considered to have completed the double-blind period until Day 181 of the study.

4.4.2.2. Long-Term Follow-Up Period

A participant in Cohort 2 will be considered to have completed the study if he or she has completed the double-blind LTFU period (Year 1 visit, Day 366).

The end of Cohort 2 is considered as the Year 1 visit (Day 366) for the last participant in Cohort 2.

The end of study is considered as the Year 5 visit (Day 1826) for the last participant in Cohort 1. The final data from the study site will be sent to the sponsor (or designee) after completion of the final participant visit at that study site, in the time frame specified in the clinical trial agreement.

5. STUDY POPULATION

Screening for eligible participants will be performed within 28 days before administration of the study vaccine. Refer to Section 5.4, Screen Failures for conditions under which the repeat of any screening procedures is allowed.

The inclusion and exclusion criteria for enrolling participants in this study are described below. If there is a question about these criteria, the investigator must consult with the appropriate sponsor representative and resolve any issues before enrolling a participant in the study. Waivers are not allowed.

5.1. Inclusion Criteria

Each potential participant must satisfy all of the following criteria to be enrolled in the study:

1. male or female, ≥60 to ≤85 years of age for Cohort 1 or ≥60 years of age for Cohort 2, inclusive, on the day of signing the informed consent form (ICF) and available for the duration of the study.

2. Criterion modified per Amendment 4.

2.1 must have a body mass index (BMI) of >18.5 to <40 kg/m^2.

3. must be healthy or medically stable, in the investigator's clinical judgment, as confirmed by medical history, physical examination, vital signs, and, for Cohort 1 only, by 12-lead electrocardiogram (ECG) and clinical laboratory tests, performed at the screening visit. Participant may have underlying illnesses such as hypertension, diabetes, or ischemic heart disease, as long as their symptoms/signs are medically controlled. If he/she is on medication for a condition, the medication dose must have been stable for at least 12 weeks preceding vaccination and expected to remain stable for the duration of the study (or minor dose adaptations could be done and accepted based on physician's judgment).

   applicable for Cohort 1 only: If laboratory screening tests are outside the normal reference ranges, repeat of screening tests is permitted once (at the discretion of the investigator) during screening to assess eligibility. Enrollment of a participant with clinical laboratory values outside of the central laboratory normal range representing toxicity Grade 1 or Grade 2 is allowed if the investigator considers the values not to be clinically significant and reasonable for the population under study. Refer to the toxicity grade assessment are provided in Section 10.5 Appendix 5 Toxicity Tables. This determination must be recorded in the participant's source documents by the investigator.

   Note: If laboratory screening tests are out of central laboratory normal ranges and deemed clinically significant, repeat of screening tests is permitted once, using an

unscheduled visit during the screening period to assess eligibility. Screening laboratory tests are to be done within 28 days of study vaccination.

4. before randomization, a woman must be:
    a. postmenopausal
    A postmenopausal state is defined as no menses for 12 months without an alternative medical cause; or
    b. not intending to conceive by any methods.
5. must sign an ICF indicating that he or she understands the purpose of, and procedures required for, the study and is willing to participate in the study.
6. willing and able to adhere to the lifestyle restrictions specified in this protocol (refer to Section 5.3).
7. agrees not to donate blood until 12 weeks after receiving the study vaccine.
8. must be willing to provide verifiable identification, has means to be contacted and to contact the investigator during the study.
9. criterion per Amendment 3, applicable for Cohort 2 only: must have a documented history of UTI in the past 5 years.

5.2. Exclusion Criteria

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

1. applicable for Cohort 1 only: must not have been previously vaccinated with Prevnar 13 (or other PCVs) and must not be planning to receive any PCV until the final analysis (Day 181) is performed. Also, vaccination with PPSV23 (PNEUMOVAX 23®) should have occurred at least one year prior to screening, and the participant is not planning to receive the PPSV23 until the final analysis (Day 181) is performed.
2. acute illness (this does not include minor illnesses such as diarrhea or mild upper respiratory tract infection) or temperature ≥38.0ºC (100.4°F) within 24 hours prior to the administration of study vaccine, or, applicable for Cohort 2 only, an ongoing or suspected symptomatic UTI; enrollment at a later date is permitted (provided the screening window of 28 days is respected).
3. history of malignancy within 5 years before screening (exceptions are squamous and basal cell carcinomas of the skin and carcinoma in situ of the cervix, or malignancy, which is considered cured with minimal risk of recurrence).
4. known allergies, hypersensitivity, or intolerance to ExPEC10V or its excipients (refer to Investigator's Brochure).[3]

5. applicable for Cohort 1 only: known allergy or history of anaphylaxis or other serious adverse reactions to vaccines or vaccine products (including any of the constituents of the active control vaccines) (refer to ExPEC4V Investigator's Brochure[2] or Prevnar 13 package insert[9]).

6. contraindication to IM injections and blood draws eg, bleeding disorders.

7. abnormal function of the immune system resulting from:
    a. Clinical conditions (eg, autoimmune disease or immunodeficiency).
    b. Chronic or recurrent use of systemic corticosteroids.
    Note: Ocular, topical or inhaled steroids are allowed.
    c. Administration of antineoplastic and immunomodulating agents or radiotherapy.

8. history of acute polyneuropathy (eg, Guillain-Barré syndrome).

9. history of chronic urticaria (recurrent hives), eczema or atopic dermatitis.

10. received treatment with immunoglobulins in the 2 months or blood products in the 4 months before the planned administration of the study vaccine or has any plans to receive such treatment during the study.

11. received or plans to receive:
    a. licensed live attenuated vaccines - within 28 days before or after planned administration of the study vaccine.
    b. other licensed (not live) vaccines - within 14 days before or after planned administration of the study vaccine.

12. received an investigational drug or used an invasive investigational medical device or received an investigational vaccine within 90 days before the planned administration of the study vaccine or is currently enrolled or plans to participate in another investigational study until Day 181 in this study. *Note*: Participation in an observational clinical study is allowed with prior approval of the sponsor. During the LTFU periods in Cohort 1 and Cohort 2, participation in another investigational study is allowed only with prior approval of the sponsor.

13. history of an underlying clinically significant acute or uncontrolled chronic medical condition or physical examination findings for which, in the opinion of the investigator, participation would not be in the best interest of the participant (eg, compromise the well-being) or that could prevent, limit, or confound the protocol-specified assessments.

14. had major surgery (per the investigator's judgment) within 4 weeks prior to randomization, or has surgery planned during the time the participant is expected to participate in the study or within 6 months after the last study vaccine administration.

    Note: Participants with planned surgical procedures to be conducted under local anesthesia may participate and not judged as major by the investigator may participate.

15. employee of the investigator or study site, with direct involvement in the proposed study or other studies under the direction of that investigator or study site, as well as family members of the employees or the investigator, or an employee of the sponsor.

16. applicable for Cohort 1 only: chronic active hepatitis B or hepatitis C infection, measured respectively by hepatitis B surface antigen test or by hepatitis C virus [HCV] antibody test; if positive, HCV RNA polymerase chain reaction test will be used to confirm active versus past HCV infection.

16.1 criterion per Amendment 3, applicable for Cohort 2 only: evidence of chronic active hepatitis B or hepatitis C infection by medical history.

17. applicable for Cohort 1 only: test positive for human immunodeficiency virus (HIV) type 1 or type 2 infection at screening.

17.1 criterion per Amendment 3, applicable for Cohort 2 only: evidence of HIV type 1 or type 2 infection by medical history.

18. has had major psychiatric illness and/or drug substance or alcohol abuse in the past 12 months which in the investigator's opinion would compromise the participant's safety or compliance with the study procedures.

19. cannot communicate reliably with the investigator.

20. who, in the opinion of the investigator, is unlikely to adhere to the requirements of the study, or is unlikely to complete the vaccination and subsequent follow-up period(s).

NOTE: Investigators should ensure that all study enrollment criteria have been met at screening. If a participant's clinical status changes (including any available laboratory results or receipt of additional medical records) after screening but before the first dose of study vaccination is given such that he or she no longer meets all eligibility criteria, then the participant should be excluded from participation in the study. Section 5.4, Screen Failures, describes options for retesting. The required source documentation to support meeting the enrollment criteria are noted in Section 10.2 Appendix 2, Regulatory, Ethical, and Study Oversight Considerations.

5.3. Lifestyle Considerations

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

1. Refer to Section 6.5, Concomitant Therapy for details regarding prohibited and restricted therapy during the study.

2. Agree to follow all requirements that must be met during the study as noted in the Inclusion and Exclusion Criteria (Section 5.1 and Section 5.2, respectively).

5.4. Screen Failures

Participant Identification, Enrollment, and Screening Logs

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

The participant identification and enrollment log will be treated as confidential and will be filed by the investigator in the study file. To ensure participant confidentiality, no copy will be made. All reports and communications relating to the study will identify participants by participant identification and age at initial informed consent. In cases where the participant is not randomized into the study, the date seen and age at initial informed consent will be used.

Rescreening

Individuals who do not meet the criteria for participation in this study due to acute illness or, applicable for Cohort 2 only, an ongoing or suspected symptomatic UTI may have a repeat assessment only once within the 28 day screening period.

Rescreened participants outside the 28 day window of screening period will be assigned a different participant number and all screening assessments are to be repeated.

6. STUDY INTERVENTION

6.1. Study Vaccines Administered

ExPEC10V (VAC52416): *E. coli* bioconjugate vaccine in phosphate buffered solution containing O-antigen PSs of ExPEC serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B and O75 separately bioconjugated to the EPA carrier protein.

Single 0.5 mL IM (deltoid) injection of one of the three doses of ExPEC10V on Day 1 (Table 7).

ExPEC10V will be supplied as follows: high dose of 176 µg PS/mL, medium dose of 120 µg PS/mL and diluent. High dose will be prepared by aliquoting of ExPEC10V 176 µg PS/mL vial, medium dose will be prepared by aliquoting of ExPEC10V 120 µg PS/mL vial and low dose (88 µg PS/mL) will be prepared by aliquoting of ExPEC10V 176 µg PS /mL vial and the diluent.

ExPEC4V (JNJ-63871860): *E. coli* bioconjugate vaccine in saline buffer solution containing O-antigen PSs of ExPEC serotypes O1A, O2, O6A, and O25B (4:4:4:8 µg PS/ExPEC serotypes) separately bioconjugated to the EPA carrier protein. ExPEC4V dose was identified from the primary analysis data of the Phase 2 study 63871860BAC2001.

Single 0.5 mL IM (deltoid) injection of ExPEC4V on Day 1.

ExPEC4V will be supplied as ExPEC4V 8:8:8:16 µg PS/mL vial and the 4:4:4:8 µg PS dose will be prepared by aliquoting of ExPEC 4V 8:8:8:16 µg PS/mL vial.

Note: ExPEC4V clinical study material will be labeled as "8/8/8/16 µg PS/mL".

Prevnar 13: Sterile suspension of saccharides of the capsular antigens of *Streptococcus pneumoniae* serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F, individually linked to non-toxic diphtheria CRM197 protein.

Single 0.5 mL IM (deltoid) injection of Prevnar 13 on Day 1, supplied in a single-dose prefilled syringe.

Placebo: Normal saline.

Single 0.5 mL IM (deltoid) injection of placebo on Day 1.

Study vaccine administration must be captured in the source documents and the electronic case report form (eCRF).

ExPEC10V will be manufactured and provided under the responsibility of the sponsor. Refer to the Investigator's Brochure for a list of excipients.

6.2. Preparation/Handling/Storage/Accountability

In Cohort 1, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation and administration. Study vaccine will be administered IM by an unblinded vaccine administrator at the study site who can be a trained and qualified study nurse, medical doctor, or otherwise qualified healthcare professional. These unblinded members will not be part of the team performing the evaluations.

In Cohort 2, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation. These unblinded members

will not be part of the team performing the evaluations. The vaccine administrator will be blinded and can perform other study evaluations.

Refer to the investigational product and procedures manual for additional guidance on study vaccine preparation, handling, and storage.

The unblinded designated pharmacist(s)/qualified staff member(s) is responsible for ensuring that all study vaccines received at the site are inventoried and accounted for throughout the study. The study vaccine administered to the participant must be documented on the vaccine accountability form. All study vaccines will be stored and disposed of according to the sponsor's instructions. Study-site personnel must not combine contents of the study vaccine containers.

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

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

6.3. Measures to Minimize Bias: Randomization and Blinding

Vaccination Allocation

Procedures for Randomization

Participants in Cohort 1 will be randomly assigned to 1 of 5 study vaccination groups (1 of 3 doses of ExPEC10V, ExPEC4V or Prevnar 13) based on a computer-generated randomization schedule prepared before the study by or under the supervision of the sponsor. Randomization will be performed in a staggered manner in different steps (see Table 1). In between the different steps, randomization will be halted until PI(s)/SRP/SML assess the 24-hour safety data of the sentinel participants or a DRC assessment of the 14-day post-vaccination safety data has been performed (see Figure 3 for details).

Participants in Cohort 2 will be randomly assigned to 1 of 2 study vaccination groups (the selected dose of ExPEC10V or placebo) based on a computer-generated randomization schedule prepared before the study by or under the supervision of the sponsor.

The interactive web response system (IWRS) will assign a unique treatment code, which will dictate the study vaccination assignment. The requestor must use his or her own user identification

and personal identification number when contacting the IWRS and will then give the relevant participant details to uniquely identify the participant.

In addition for Cohort 2, 33% of the participants will be randomly selected using the IWRS on Day 1 to provide a stool sample at the Day 30 and 181 visits (refer to Section 8.5, Other Evaluations).

Blinding

The investigator will not be provided with randomization codes. The codes will be maintained within the IWRS, which has the functionality to allow the investigator to break the blind for an individual participant.

Data that may potentially unblind the study vaccination assignment (ie, different packaging of the study vaccines as well as preparation) will be handled with special care to ensure that the integrity of the blind is maintained and the potential for bias is minimized.

In Cohort 1, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation and administration. Study vaccine will be administered IM by an unblinded vaccine administrator at the study site who can be a trained and qualified study nurse, medical doctor, or otherwise qualified healthcare professional. These unblinded members will not be part of the team performing the evaluations.

Under normal circumstances, the blind should not be broken until all participants have completed the observer-blind period and the database for final analysis is locked. After final analysis database lock, the study participants will be informed by a telephone contact whether that telephone contact will be their last study visit or whether they will be progressing to the LTFU period. The LTFU period for Cohort 1 is open-label as participants not included in the Prevnar 13 group will be offered Prevnar 13 vaccination outside of the study after the final analysis database lock (Day 181).

In Cohort 2, the participants, clinical staff, investigators, and sponsor personnel will be blinded to study vaccination group allocation, except for the designated pharmacist(s) or qualified staff member(s) with primary responsibility for study vaccine preparation. These unblinded members will not be part of the team performing the evaluations. The vaccine administrator will be blinded and can perform other study evaluations. The blind should not be broken throughout the study until all participants have completed the double-blind LTFU period. The sponsor will be blinded to study vaccination group allocation until the database lock for the primary analysis.

Otherwise, in both cohorts of the study, the blind should be broken only if specific emergency treatment/course of action would be dictated by knowing the study vaccination group status of the participant. In such cases, the investigator may in an emergency determine the identity of the study vaccination by contacting the IWRS. It is recommended that the investigator contacts the sponsor or its designee if possible to discuss the particular situation, before breaking the blind. Telephone contact with the sponsor or its designee will be available 24 hours per day, 7 days per week. In the

event the blind is broken, the sponsor must be informed as soon as possible. The date and reason for the unblinding must be documented by the IWRS, in the appropriate section of the eCRF and in the source document. The documentation received from the IWRS indicating the code break must be retained with the participant's source documents in a secure manner.

Participants who have had their study vaccination assignment unblinded should continue to return for scheduled evaluations.

In general, randomization codes will be disclosed fully only if the study is completed and the clinical database is closed. However, for the primary analysis, the randomization codes and, if required, the translation of randomization codes into vaccination and control groups will be disclosed to those authorized sponsor personnel involved in the analysis of the data. At the time of the primary analysis, the participants, investigator(s) and site staff will remain blinded to individual participants' study vaccination assignment.

6.4. Study Vaccination Compliance

In Cohort 1, study vaccine will be administered as an IM injection by an unblinded vaccine administrator at the study site who can be a trained and qualified study nurse, medical doctor, or otherwise qualified healthcare professional and will not be part of the team performing the evaluations.

In Cohort 2, study vaccine will be administered as an IM injection by a blinded vaccine administrator at the study site, who can perform other study evaluations.

The date and time of the study vaccine administration and deltoid used for injection will be recorded in the eCRF.

6.5. Concomitant Therapy

Prestudy specific therapies (such as non-steroidal anti-inflammatory drugs, corticosteroids, antihistaminic, vaccinations) administered up to 30 days before first dose of study vaccine must be recorded at screening.

Concomitant therapies (such as non-steroidal anti-inflammatory drugs, corticosteroids, antihistaminic, vaccinations) must be recorded for all participants from the signing of the ICF until 29 days after the study vaccination. Use of oral licensed *E. coli* vaccines (eg, Uro-Vaxom [OM-89] or Uromune) is allowed for the duration of the study, including long-term follow-up, and must always be recorded as concomitant therapy (applicable to European Union countries only).

Use of any investigational medication (including investigational vaccines other than the study vaccine) within 90 days before vaccination in the study or until Day 181 in the study is not allowed. During the LTFU periods in Cohort 1 and Cohort 2, participation in another investigational study is allowed only with prior approval of the sponsor.

A participant in Cohort 1 must not have been previously vaccinated with Prevnar 13 (or other PCV) and must not be planning to receive any PCV until the final analysis (Day 181) is performed. Also, vaccination with PPSV23 (PNEUMOVAX 23) should have occurred at least one year prior to screening, and the participant is not planning to receive the PPSV23 until the final analysis (Day 181) is performed.

Vaccination with licensed live attenuated vaccines within 28 days of a study vaccination (ie, before and after) is prohibited. Other licensed vaccines (not live) should be given at least 14 days before (or at least 14 days after) administration of study vaccine in order to avoid potential confusion of adverse reactions and potential immune interference. If a vaccine is indicated in a post-exposure setting (eg, rabies or tetanus), it must take priority over the study vaccination.

Vaccination with an authorized/licensed COVID-19 vaccine (eg, vaccination for COVID-19 through Emergency Use Authorization) is permitted when given at least 28 days before or after planned administration of the study vaccination. Use of COVID-19 vaccines must always be recorded as concomitant therapy for the entire duration of the study.

Chronic or recurrent use of immunomodulators/suppressors, eg, cancer chemotherapeutic agents, systemic corticosteroids is prohibited during the study. Note: Ocular, topical or inhaled steroids are allowed.

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

6.6. Dose Modification

Not applicable.

6.7. Intervention After the End of the Study

Participants in Cohort 1 randomized to either of the three doses of ExPEC10V as well as ExPEC4V participants (Groups 1-4; see Table 1) will be offered Prevnar 13 vaccination outside of the study after the final analysis database lock (Day 181) in order to follow ACIP recommendations published in September 2014.[1]

7. DISCONTINUATION OF STUDY INTERVENTION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1. Discontinuation of Study Vaccination

Since this is a single vaccination study, this section is not applicable.

7.1.1. Contraindications to Study Vaccination

The following events constitute a contraindication to vaccination on Day 1:

- Acute illness or acute infection within 24 hours prior to the administration of study vaccine (this does not include minor illnesses such as mild diarrhea or mild upper respiratory tract infection).
- Fever (temperature ≥38.0°C [100.4°F]) within 24 hours prior to the administration of study vaccine.
- Applicable for Cohort 2 only: ongoing or suspected symptomatic UTI.

If any of these events occur within 24 hours prior to the vaccination on Day 1, then enrollment at a later date is permitted (provided the screening window of 28 days is respected). If outside the 28 day screening window and the participant fulfills all inclusion and exclusion criteria (Section 5, Study Population), the participant might be reconsented, rescreened (under new participant number), enrolled, and vaccinated after the event(s) has resolved (provided the enrollment will be ongoing).

7.2. Participant Discontinuation/Withdrawal From the Study

Each participant has the right to withdraw from the study at any time for any reason without affecting the right to treatment by the investigator. Although the participant is not obliged to give reason(s) for withdrawing prematurely, the investigator should make a reasonable effort to ascertain the reason(s) while fully respecting the participant's rights.

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

- Lost to follow-up
- Withdrawal of consent
- Death
- Noncompliance defined as failed to receive the study vaccine
- Repeated failure to comply with protocol requirements
- Decision by the sponsor to stop or cancel the study
- Decision by the investigator to withdraw participants
- Decision by local regulatory authorities and Institutional Review Board/Independent Ethics Committee (IRB/IEC) to stop or cancel the study

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

When a participant withdraws before completing the study, the reason for withdrawal is to be documented in the eCRF and in the source document. Study vaccination assigned to the withdrawn

participant may not be assigned to another participant. Additional participants will be entered provided the enrollment will be ongoing. If a participant withdraws early from the study, assessments for early withdrawal should be obtained (see Early Withdrawal/Exit Visit in Section 8. Study Assessments and Procedures). Participants who wish to withdraw consent from participation in the study will be offered a single exit visit (see Early Withdrawal/Exit Visit in Section 8. Study Assessments and Procedures) for safety follow-up (prior to formal withdrawal of consent). They have the right to refuse.

7.2.1. Withdrawal From the Use of Research Samples

Withdrawal From the Use of Samples in Future Research

The participant may withdraw consent for use of samples for research (refer to Long-Term Retention of Samples for Additional Future Research in Section 10.2 Appendix 2, Regulatory, Ethical, and Study Oversight Considerations). In such a case, samples will be destroyed after they are no longer needed for the clinical study. Details of the sample retention for research are presented in the main ICF.

7.3. Lost to Follow-up

If a participant is lost to follow-up, every reasonable effort must be made by the study-site personnel to contact the participant and determine the reason for discontinuation/withdrawal. The measures taken to follow-up must be documented. Refer to Section 7.2, Participant Discontinuation/Withdrawal From the Study.

8. STUDY ASSESSMENTS AND PROCEDURES

The overview of study assessments and procedures will be the same for both cohorts in the study, unless stated otherwise.

Overview

Section 1.3 Schedule of Activities summarizes the frequency and timing of safety and immunogenicity measurements applicable to this study.

After vaccination, participants will remain under observation at the study site for at least 30 minutes for presence of any acute reactions and solicited events or longer if deemed necessary by the investigator. The participant's vital signs will also be collected after 30 minutes post-vaccination. Any unsolicited AEs and solicited local or systemic AEs will be documented by study-site personnel following this observation period. Participants will be provided with a thermometer (to measure body temperature), ruler (to measure local injection-site reactions) and daily assessment (participant) ediary with instructions for the proper recording of events.

The ediary includes instructions on how to capture the data and grading scales to assess severity of the symptoms. The study staff is responsible for providing appropriate training to the participant to avoid missing or incorrect data (refer to Study Training Manual). The ediary will be reviewed by the study personnel at visits indicated in the Section 1.3 Schedule of Activities. If the ediary

review is missed, the ediary will be reviewed at the following visit. If any participant experiences any issue with the ediary entry at any time in between vaccination (Day 1) and 14 days post-vaccination (Day 15) visit, an optional telephone contact(s) should be made to collect safety data.

Each participant will record solicited local (at injection site) and systemic AEs and body temperatures (oral route preferred), beginning in the evening of the day of study vaccination and on a daily basis for the following 14 days. Body temperatures (oral route preferred) should be taken at approximately the same time each day.[a] If more than one measurement is made on any given day, the highest temperature of that day will be used in the eCRF. Study-site personnel will collect and review participant ediary information and confirm the entries at subsequent site visits.

In Phase 1 of Cohort 1, all (12) sentinel participants (four participants at each dose level) will be contacted by telephone 24 hours post-vaccination to collect safety information by study-site personnel. All participants will be contacted by telephone 48 hours post-vaccination. The participants will also be instructed to contact the site in case of any signs or symptoms.

All pre-existing medical conditions until the time of vaccination should be recorded as medical history.

Solicited AEs will be collected from the time of vaccination through 14 days after the vaccination (Day 1 to Day 15). Unsolicited AEs will be collected from the time of vaccination through 29 days after the vaccination (Day 1 to Day 30). All AEs and special reporting situations, whether serious or non-serious, that are related to study procedures or that are related to non-investigational (concomitant) Janssen products will be reported from the time a signed and dated ICF is obtained onwards. All other AEs, SAEs and special reporting situations, will be reported from the day of first vaccination onwards until Day 181 of the study. In the LTFU periods in Cohort 1 and Cohort 2, only SAEs related to the study vaccine or study procedures will be collected.

All AEs, including any that are ongoing at 29 days after vaccination, will be followed until clinical resolution or stabilization. Concomitant therapies will be collected from the vaccination until 29 days and additionally outside of these periods when associated with an SAE.

For Cohort 1, blood will be collected for laboratory safety assessments at the screening visit, on Day 1 (pre-vaccination), Day 15 and Day 30 (and at the exit visit if a participant terminates before the end of Day 30 without withdrawing consent[b]).

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[a] Temperature 14 days after vaccination (Day 15) may be collected earlier to coincide with the clinic visit.

[b] Blood samples for safety laboratory (biochemistry and hematology) will only be taken if the early exit visit is within 29 days of the vaccination.

Blood samples of approximately 10 mL will be collected from all participants to assess immunogenicity pre-vaccination on the day of vaccination (Day 1), on Days 15, 30 and 181 (or at the early exit visit if the participant prematurely terminates the study before Day 181 without withdrawing consent). Additional blood samples will be collected from participants in the ExPEC10V selected dose group and the Prevnar 13 group in Cohort 1 at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826) and from all participants in Cohort 2 at Year 1 post-vaccination. In Cohort 1, at the Day 30 visit, an additional 50 mL blood sample will be drawn from a total of 30 participants willing to donate blood, for development or validation of the immunological assays (multiplex ECL-based immunoassay and MOPA) and for assessment of the validity and consistency of each assay experiment. This additional blood draw will be collected in a stepwise manner:

- Up to 15 participants from low and medium dose groups in Phase 1
- Up to 15 participants from high dose group in Phase 1
- If needed, remainder from Phase 2a participants

The timings of the post-vaccination visits will be determined relative to the actual day of the corresponding vaccination.

For Cohort 2, a baseline stool sample will be collected from all participants. At randomization on Day 1, a subset of 33% of participants will be randomly selected (using the IWRS) to provide stool samples at the Day 30 and 181 visits. Stool samples will be analyzed to evaluate the effect of ExPEC10V on the prevalence of pathogens (eg, *Clostridium Difficile*) and ExPEC10V serotypes in the intestinal flora using metagenomics.

Study Visits in Cohort 1 - Observer-Blind Period (All Participants)

- **Screening (Visit 1)** will be performed within 28 days prior to vaccination, and will include: written informed consent, inclusion/exclusion criteria, demographics, 12-lead ECG, vital signs[a] including body temperature, height and weight, medical history, prestudy and concomitant medications, full physical examination, blood samples for HIV-1/2 and hepatitis B/C serology and for clinical laboratory testing. Screening laboratory tests are to be done within 28 days prior to the administration of study vaccine. The screening visit may be split over several days.

  For entry into the study, each participant must be healthy on the basis of clinical laboratory tests performed at screening. Participants with laboratory values or vital signs (eg, elevated blood pressure) not meeting eligibility criteria at the screening visit may have one repeat testing at the discretion of the investigator if the abnormality is not clinically significant or may be a testing aberrancy.

[a] Supine systolic and diastolic blood pressure, pulse/heart rate and respiratory rate after at least 5 minutes rest.

Enrollment of a participant with clinical laboratory values outside of the central laboratory normal range representing toxicity Grade 1 or Grade 2 is allowed if the investigator considers the values not to be clinically significant and reasonable for the population under study. Refer to the toxicity grade assessment are provided in Section 10.5 Appendix 5 Toxicity Tables.

- **Study Day 1 (Visit 2):**

  pre-vaccination: recheck inclusion/exclusion criteria, check for medical history, vital signs (pre-vaccination), targeted physical examination, pre-vaccination symptoms, blood samples for immunogenicity and for clinical laboratory testing, followed by randomization in the study.

  post-vaccination: administration of study vaccine (observe for 30 minutes post-vaccination), vital signs (30 minutes post-vaccination), recording of solicited and unsolicited AEs, SAEs and concomitant medications, and ediary distribution.

  Enrollment and vaccination of participants will be done in a staggered approach following the stepwise dose-escalation procedures to allow for safety review of four sentinel participants at 24 hours after vaccination followed by a 14-day post-vaccination safety data review of 28 participants by DRC at each dose level (first at low dose, second at medium dose followed by high dose).

- **Study Day 2 (Telephone Contact)** 24 hours post-vaccination: only sentinel participants (12) will be contacted by telephone 24 hours post-vaccination to collect safety information, recording of solicited and unsolicited AEs, SAEs and concomitant medications.

- **Study Day 3 (Telephone Contact)** 2 days/+1 day post-vaccination: recording of solicited and unsolicited AEs, SAEs and concomitant medications.

- **Study Day 15 (Visit 3)** 14 days/±2 days post-vaccination: vital signs, targeted physical examination, review and collection of ediary, recording of solicited and unsolicited AEs, SAEs and concomitant medications, collection of blood for clinical laboratory testing and immunogenicity assessment. If the visit occurs before the end of the diary period, review of the ediary will still take place, but the ediary will be returned by the participant at the next study visit (Day 30).

  Note: If any of the first 84 participants come in earlier than Day 15 for Visit 3 (allowed window is ±2 days), a subsequent phone call will be made at the end of the diary period to collect ediary information recorded between the actual visit and the end of the diary period on Day 15 which will be captured in the eCRF for DRC review.

- **Study Day 30 (Visit 4)** 29 days/±3 days post-vaccination: vital signs, targeted physical examination, recording of unsolicited AEs, SAEs and concomitant medications, collection of blood for clinical laboratory testing and immunogenicity assessment.

An additional 50 mL immunogenicity blood sample will also be collected from a total of 30 participants willing to donate blood for development or validation of assays. This additional blood draw will be collected in a stepwise manner from the participants, initially up to 15 participants from low and medium dose groups in Phase 1; then up to 15 participants from high dose group in Phase 1 and if needed, remainder from Phase 2a participants.

- **Study Day 181 (Visit 5)** 6 months/±14 days post-vaccination: targeted physical examination, vital signs, collection of SAEs and concomitant medications only in conjunction with SAEs. Blood samples will be taken for immunogenicity assessment.
- **Early Exit Visit**: targeted physical examination, vital signs, recording of solicited AEs and ediary (if within 14 days post-vaccination), unsolicited AEs (if within 29 days post-vaccination), collection of SAEs and concomitant medications (if occurring after Day 30, then concomitant medications only in conjunction with SAEs). Blood samples will be taken for immunogenicity assessment. If early exit visit occurs before 29 days post-vaccination, then blood samples will also be taken for safety assessment.

Study Visits in Cohort 2 - Double-Blind Period Until Day 181 (All Participants)

- **Screening (Visit 1)** will be performed within 28 days prior to vaccination or on the day of vaccination, and will include: written informed consent, inclusion/exclusion criteria, demographics, vital signs[a] including body temperature, height and weight, medical history, prestudy and concomitant medications, and full physical examination. The screening visit may be split over several days. If screening is performed on the day of vaccination, Visit 1 and Visit 2 will coincide on Day 1. Screening must be completed, and all eligibility criteria must be fulfilled prior to randomization and vaccination.

  For entry into the study, each participant must be healthy or medically stable. Participants with vital signs (eg, elevated blood pressure) not meeting eligibility criteria at the screening visit may have one repeat testing at the discretion of the investigator if the abnormality is not clinically significant or may be a testing aberrancy.

  All participants will be provided with a stool sample kit to collect a baseline stool sample, which should be available before vaccination. Baseline stool samples should be collected at home by each participant between 1 and 7 days prior to the scheduled Day 1 visit; stool samples should be provided to the study-site personnel upon arrival on Day 1.

[a] Supine systolic and diastolic blood pressure, pulse/heart rate and respiratory rate after at least 5 minutes rest.

- **Study Day 1 (Visit 2)**:

  pre-vaccination: recheck inclusion/exclusion criteria, check for medical history, vital signs (pre-vaccination), targeted physical examination, pre-vaccination symptoms, blood samples for immunogenicity, followed by randomization in the study. Collection of stool sample from all participants.

  post-vaccination: administration of study vaccine (observe for 30 minutes post-vaccination), vital signs (30 minutes post-vaccination), recording of solicited and unsolicited AEs, SAEs and concomitant medications, and ediary distribution.

- **Study Day 3 (Telephone Contact)** 2 days/+1 day post-vaccination: recording of solicited and unsolicited AEs, SAEs, and concomitant medications.

- **Study Day 15 (Visit 3)** 14 days/±2 days post-vaccination: vital signs, targeted physical examination, review and collection of ediary, recording of solicited and unsolicited AEs, SAEs and concomitant medications, and collection of blood for immunogenicity assessment. If the visit occurs before the end of the ediary period, review of the ediary will still take place, but the ediary will be returned by the participant at the next study visit (Day 30). The subset of participants selected for stool sample analysis at randomization on Day 1 will be provided with a stool sample kit for collection of the Day 30 stool sample.

- **Study Day 30 (Visit 4)** 29 days/±3 days post-vaccination: vital signs, targeted physical examination, recording of unsolicited AEs, SAEs, and concomitant medications, collection of blood for immunogenicity assessment. Collection of stool sample from the subset of participants selected at randomization on Day 1. The sample should be collected by the participant between 1 and 7 days prior to the study visit and provided to the study-site personnel upon arrival for the visit. If the participant does not bring the stool sample to the visit, he or she may deliver the stool sample to the study site up to 3 days after the visit. The selected participants will be provided with a stool sample kit for collection of the Day 181 stool sample.

- **Study Day 181 (Visit 5)** 6 months/±14 days post-vaccination: targeted physical examination, vital signs, collection of SAEs and concomitant medications only in conjunction with SAEs. Blood samples will be taken for immunogenicity assessment. Collection of stool sample from the subset of participants selected at randomization on Day 1. The sample should be collected by the participant between 1 and 7 days prior to the study visit and provided to the study-site personnel upon arrival for the visit. If the participant does not bring the stool sample to the visit, he or she may deliver the stool sample to the study site up to 3 days after the visit.

- **Early Exit Visit**: targeted physical examination, vital signs, recording of solicited AEs and ediary (if within 14 days post-vaccination), unsolicited AEs (if within 29 days post-vaccination), collection of SAEs and concomitant medications (if occurring after Day 30, then concomitant medications only in conjunction with SAEs). Blood samples will be taken for immunogenicity assessment.

Study Visits in Cohort 1 - Open-Label LTFU Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 - Double-Blind LTFU Period (All Participants)

For all participants in Cohort 1 of the study, a telephone contact will be scheduled (after final analysis database lock, Day 181) to inform them whether that telephone contact will be their last

study visit or whether they will be progressing to the LTFU period. All participants in Cohort 2 will progress to the double-blind LTFU period.

- **Study Day 366 (Year 1, Visit 6)** 12 months/±14 days post-vaccination: In Cohort 1, reconsent from all participants will be obtained before entering the open-label LTFU period. Collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment. This is the only LTFU visit for Cohort 2 participants. Participants in Cohort 2 who have already completed their Year 1 LTFU visit at the time of implementation of protocol amendment 8, will receive a safety follow-up telephone call, informing them they have completed the study due to the implementation of a protocol amendment. This telephone call should be performed as soon as possible, and before the cancelled Year 2 LTFU visit that was planned to be performed under the original protocol.
- **Study Day 731 (Year 2, Visit 7)** 24 months/±28 days post-vaccination: collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment. This LTFU visit applies to Cohort 1 participants only.
- **Study Day 1096 (Year 3, Visit 8)** 36 months/±28 days post-vaccination: collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment. This LTFU visit applies to Cohort 1 participants only.
- **Study Day 1461 (Year 4, Visit 9)** 48 months/±28 days post-vaccination: collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment. This LTFU visit applies to Cohort 1 participants only.
- **Study Day 1826 (Year 5, Visit 10)** 60 months/±28 days post-vaccination: collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment. This LTFU visit applies to Cohort 1 participants only.
- **Early Exit Visit**: collection of SAEs related to the study vaccine or study procedures and concomitant medications in conjunction with these SAEs. Blood samples will be taken for immunogenicity assessment, except when the exit visit is replaced by a telephone contact (see below).

Participants in Cohort 1 will need to provide their reconsent for the Year 4 and Year 5 safety and immunogenicity LTFU visits, the latest just before the Year 4 visit. Participants in Cohort 1 who do not provide their reconsent for the Year 4 and Year 5 LTFU visits will be seen as early withdrawal.

Participants who are unable to continue participation in the study, but who do not withdraw consent, should be encouraged to complete an exit visit, at the investigator's discretion. If the exit visit would occur shortly after an annual visit, the investigator may decide to have the exit visit replaced by a telephone contact.

In Cohort 1, the total blood volume for the study to be collected from each participant (except the 30 participants, see below) will be approximately 120 mL (30 mL for safety and 90 mL for

immunogenicity [40 mL in observer-blind period and 50 mL in LTFU period]). In Cohort 1, on Day 30, an additional 50 mL blood sample will also be collected from a total of 30 participants willing to donate blood. This additional blood draw will be collected in a stepwise manner from participants, initially up to 15 participants from low and medium dose groups in Phase 1; followed by up to 15 participants from high dose group in Phase 1 and if needed, remainder from Phase 2a participants.

In Cohort 2, the total blood volume for the study to be collected from each participant will be approximately 50 mL (for immunogenicity; 40 mL in double-blind period until Day 181 and 10 mL in LTFU period).

The maximum amount of blood drawn from each participant in this study will not exceed 550 mL in any 8-week period (per US Department of Health and Human Services [HHS] Office for Human Research Protections [OHRP], and US FDA guidelines).[5,10] Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

Sample Collection and Handling

The actual dates and times of sample collection must be recorded in the eCRF or laboratory requisition form.

Refer to the Section 1.3 Schedule of Activities for the timing and frequency of all sample collections.

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

Study-Specific Materials

The investigator will be provided with the following supplies:

- Investigator's Brochures for ExPEC10V and ExPEC4V
- Package Insert for Prevnar 13
- Study site investigational product and procedures manual
- Laboratory manual
- Electronic Data Capture (eDC) Manual/ eCRF completion guidelines and randomization instructions
- IWRS manual
- Sample ICF
- Participant diaries
- Rulers and thermometers

- Participant wallet cards

8.1. Safety Assessments

Details regarding the DRC are provided in Committees Structure in Section 10.2 Appendix 2, Regulatory, Ethical, and Study Oversight Considerations.

Adverse events will be reported and followed by the investigator as specified in Section 8.2, Adverse Events and Serious Adverse Events and Section 10.3 Appendix 3, Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-Up, and Reporting.

Any clinically relevant changes occurring during the study must be recorded on the Adverse Event section of the eCRF. Any clinically significant abnormalities persisting at the end of the study/early withdrawal will be followed by the investigator until resolution or until a clinically stable condition is reached.

The study will include the following evaluations of safety and tolerability according to the time points provided in Section 1.3 Schedule of Activities:

8.1.1. Physical Examination

A full physical examination including body weight and height will be carried out at screening. At all other visits, a targeted (abbreviated and symptom-directed) examination will be performed by the investigator based on clinically relevant issues, clinically relevant symptoms and medical history. The targeted physical examination may be repeated if deemed necessary by the investigator.

Physical examinations will be performed by the investigator or an appropriately trained and qualified delegate. Any clinically relevant abnormalities or changes in severity observed during the review of body systems should be documented in the eCRF as an AE (if pre-vaccination, then as medical history).

8.1.2. Vital Signs

Blood pressure, respiratory rate, and pulse/heart rate measurements should be preceded by at least 5 minutes of rest in a quiet setting without distractions (eg, television, cell phones).

Vital signs are to be measured before blood draws and will be performed at screening, on Day 1 (pre- and 30 minutes post-vaccination), Day 15, Day 30, and Day 181, or early exit.

The following measurements will be performed:

- Pulse/heart rate (beats per minutes), respiratory rate (breaths per minute), supine systolic blood pressure (mmHg) and supine diastolic blood pressure (mmHg)
- Body temperature (oral route preferred, or in accordance with the local standard of care)

Confirmatory vital signs measurement can be done if inconsistent with a prior measurement. If any clinically significant changes in vital signs are observed, they will be reported as an AE and followed to resolution, or until reaching a clinically stable endpoint. If any clinically significant changes in vital signs are observed pre-vaccination, they will be reported as medical history.

8.1.3. Electrocardiogram (ECG) (Cohort 1)

In Cohort 1, a supine 12-lead ECG will be performed at screening and interpreted locally; ECGs will only be performed thereafter during the study if clinically indicated based on signs and symptoms.

For 30 minutes prior to the ECG, participants should refrain from meals, hot or cold beverages and strenuous exercise, and should remain in a room with a comfortable temperature. During the collection of ECGs, participants should be in a quiet setting without distractions (eg, television, cell phones). Participants should rest in a supine position for at least 5 minutes before ECG collection and should refrain from talking or moving arms or legs. If blood sampling or vital sign measurement is scheduled for the same time point as ECG recording, the procedures should be performed in the following order: ECG(s), vital signs, blood draw.

Enrollment of a participant is allowed even with abnormal ECG results as long as the investigator feels that these are not clinically significant and appropriate for the population.

8.1.4. Clinical Safety Laboratory Assessments (Cohort 1)

In Cohort 1, blood samples for serum chemistry and hematology will be collected as noted below. The investigator must review the laboratory results, document this review, and record any clinically relevant changes occurring during the study in the Adverse Event section of the eCRF. The laboratory reports must be filed with the source documents.

Laboratory Safety Assessments:

The following laboratory safety assessments will be performed at screening, on Day 1 pre-vaccination, Day 15 and Day 30, or early exit (if occurring before Day 30):

- **Blood Chemistry:**

  Sodium
  Potassium
  Creatinine
  Blood urea nitrogen (BUN)
  Aspartate aminotransferase (AST)
  Alanine aminotransferase (ALT)

- **Hematology:**

  Hemoglobin
  White blood cell (WBC) count with differential
  Platelet count

The following safety laboratory assessment will be performed only at screening:

- Serology for HIV-1/2 and for hepatitis B and C

Reporting Laboratory Abnormalities as Adverse Events

Any clinically significant abnormal laboratory value within 29 days post-vaccination that falls outside of the central laboratory normal range and that requires follow-up will be captured as an AE. Laboratory values outside normal ranges that are not clinically significant in the judgment of the investigator, should not be recorded as an AE. Any laboratory value falling within the central laboratory normal range will not be severity graded or recorded as an AE, regardless of whether the value falls within FDA ranges for Grade 1 or higher.

Repeat of Clinically Significant Laboratory Tests

For any clinically significant abnormal laboratory value that has increased in grade over baseline, the test must be repeated at the next scheduled visit or sooner based on investigator's judgment, however Grade 3 abnormalities should be retested within 48 hours. Any clinically significant abnormalities (including those persisting at the end of the study or early withdrawal) will be followed by the investigator until resolution or a clinically stable endpoint is reached.

8.2. Adverse Events and Serious Adverse Events

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

Solicited (Local and Systemic) Adverse Events

Solicited (local and systemic) AEs are precisely defined events that participants are specifically asked about and which are noted by participants through the participant ediary. All participants will be provided with an ediary and instructions on how to complete the ediary. The investigator will review each participant's ediary at the subsequent in-clinic visit; ediary information will be transcribed by the study personnel into the eCRF. In addition, after vaccination, participants will remain under observation at the study site for at least 30 minutes for presence of any acute reactions and solicited events. By definition, all solicited local AEs occurring at the vaccination site will be considered related to the study vaccine administration (injection-site reactions); relatedness of solicited systemic AEs should be determined by the investigator.

Solicited Local Adverse Events (Injection-Site Reactions)

Participants will be instructed on how to note occurrences of pain/tenderness, erythema, and swelling at the injection site daily for 14 days after vaccination (day of vaccination and the subsequent 14 days) in the participant ediary. Participants will be instructed on how to measure (using the ruler supplied) and record erythema and swelling.

- **Injection-Site Pain/Tenderness**

  Injection-site pain (eg, stinging, burning) is an unpleasant sensory and emotional experience associated with actual or potential tissue damage and occurring at the immunization site (with or without involvement of surrounding tissue). Injection-site tenderness is a painful sensation localized at the injection site upon palpation and/or movement of the limb. Due to subjective nature of the reaction, the severity assessment of pain/tenderness is self-reported (if a participant is unable to provide self-report, other reporters include parent/care giver or health care provider).[16]

- **Injection-Site Erythema**

  Injection-site erythema is a redness of the skin caused by dilatation and congestion of the capillaries localized at the injection site. It can best be described by looking and measuring.

- **Injection-Site Swelling**

  Injection-site swelling is a visible enlargement of an injected limb. It may be either soft (typically) or firm (less typical).

  Note: any other injection site events not meeting the above case definitions should be reported separately as unsolicited AEs.[19,20]

Solicited Systemic Adverse Events

Participants will be instructed on how to note daily symptoms in the participant ediary for 14 days after vaccination (day of vaccination and the subsequent 14 days), of the following systemic events: headache, fatigue, nausea, and myalgia.

Participants will also be instructed on how to record daily temperature using a thermometer provided for home use. Participants should record oral temperature in the evening after vaccination (Day 1), and then daily for the next 14 days in the participant ediary. Temperature should be measured at the same time each day. On Day 15, the temperature can be measured at the site visit (regardless of the timing). If more than one measurement is made on any given day, the highest daily temperature will be used. Fever will be recorded by the investigator in the eCRF for temperatures equal to or higher than 38.0°C (100.4°F).

Fever is defined as endogenous elevation of body temperature ≥38°C, as recorded in at least one measurement.[23]

If a solicited local or systemic AE is not resolved within 14 days after vaccination (Day 15), safety follow-up should be performed until the symptom resolves. The study-site personnel will be instructed to record the date of last symptoms and maximum severity until resolution in the source document and eCRF.

Unsolicited Adverse Events

Unsolicited AEs are all AEs for which the participant is not specifically questioned in the participant ediary.

For further details on AEs and SAEs (Definitions and Classifications; Attribution Definitions; Severity Criteria; Special Reporting Situations; Procedures) as well as product quality complaints, refer to Section 10.3 Appendix 3, Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-Up, and Reporting.

8.2.1. Time Period and Frequency for Collecting Adverse Event and Serious Adverse Event Information

All Adverse Events

All AEs and special reporting situations, whether serious or non-serious, that are related to study-related procedures or that are related to non-investigational (concomitant) sponsor products will be reported from the time a signed and dated ICF is obtained onwards. All other AEs and special reporting situations, whether serious or non-serious, will be reported from after the first vaccination onwards. Clinically relevant medical events occurring between signing of ICF and date of first vaccination will be collected on the Medical History eCRF page as pre-existing conditions.

Solicited Adverse Events

After vaccination, participants will remain under observation at the study site for at least 30 minutes for presence of any acute reactions and solicited AEs. In addition, all participants will be asked to record symptoms of the following AEs via the ediary:

- Solicited local AEs: pain/tenderness, erythema, and swelling at the injection site.

- Solicited systemic AEs: oral body temperature ≥38.0°C (100.4°F; fever), headache, fatigue, nausea, and myalgia.

Solicited local and systemic AEs will be collected for 14 days post-vaccination (Day 1 to Day 15) via the ediary, to be completed by the participants. If any participant experiences any issue with the ediary entry at any time in between vaccination (Day 1) and 14 days post-vaccination (Day 15) visit, an optional telephone contact(s) should be made to collect safety data. The investigator will review each participant's ediary at the subsequent in-clinic visit; ediary information will be transcribed by the study personnel into the eCRF. Fever will be recorded by the investigator for temperatures equal to or higher than 38.0°C (or 100.4°F). By definition, all solicited local AEs occurring at the vaccination site will be considered related to the study vaccine administration (injection-site reactions); relatedness of solicited systemic AEs should be determined by the investigator.

Unsolicited Adverse Events:

Unsolicited AEs will be collected from the administration of the study vaccine until 29 days after the study vaccination (Day 1 to Day 30) in all participants. Relatedness of unsolicited AEs should be determined by the investigator.

Serious Adverse Events

All SAEs will be collected for all participants from the administration of the study vaccine until Day 181 of the study. Additionally, AEs or SAEs that are related to the study procedures or are related to non-investigational (concomitant) Janssen products will be reported from the time a signed and dated ICF is obtained until Day 181. In the LTFU periods in Cohort 1 and Cohort 2, only SAEs related to the study vaccine or study procedures (ie, blood draws) will be collected.

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

8.2.2. Method of Detecting Adverse Events and Serious Adverse Events

Care will be taken not to introduce bias when detecting AEs or SAEs. Open-ended and nonleading verbal questioning of the participant is the preferred method to inquire about AE occurrence.

Solicited Adverse Events

Solicited AEs are predefined local at the injection site and systemic events for which the participant is specifically questioned, and which are noted by participants in their ediary (see Section 8, Study Assessments and Procedures).

Unsolicited Adverse Events

Unsolicited AEs are all AEs for which the participant is not specifically questioned in the participant ediary.

8.2.3. Follow-up of Adverse Events and Serious Adverse Events

Adverse events, including pregnancies in partners of male participants, will be followed by the investigator as specified in Section 10.3 Appendix 3, Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting.

8.2.4. Regulatory Reporting Requirements for Serious Adverse Events

The sponsor assumes the responsibility for appropriate reporting of AEs to the regulatory authorities. The sponsor will also report to the investigator (and the head of the investigational institute where required) all suspected unexpected serious adverse reactions (SUSARs). The investigator (or sponsor where required) must report SUSARs to the appropriate IEC/IRB that approved the protocol unless otherwise required and documented by the IEC/IRB. A SUSAR will be reported to regulatory authorities unblinded. Participating investigators and IEC/IRB will receive a blinded SUSAR summary, unless otherwise specified.

8.2.5. Pregnancy

All initial reports of pregnancy in partners of male participants must be reported to the sponsor by the study-site personnel within 24 hours of their knowledge of the event using the appropriate pregnancy notification form. Abnormal pregnancy outcomes (eg, spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs and must be reported using the Serious Adverse Event Form. If a participant becomes pregnant during the study, a determination regarding study vaccination discontinuation must be made by the investigator in consultation with the sponsor.

8.3. Treatment of Overdose

For this study, any dose of ExPEC10V more than the single administration (0.5 mL) of the study vaccine will be considered an overdose. For ExPEC4V and Prevnar 13, any dose more than the single administration (0.5 mL) of these vaccines will be considered an overdose. The sponsor does not recommend specific intervention for an overdose.

In the event of an overdose, the investigator or treating physician should:

- Contact the Medical Monitor immediately.
- Closely monitor the participant for AE/SAE and, for Cohort 1 only, for laboratory abnormalities until resolution.
- Document the quantity of the excess dose as well as the duration of the overdosing in the eCRF.
- Report as a special reporting situation.

8.4. Immunogenicity Assessments

Blood samples of approximately 10 mL will be collected from all participants for measurements of serum antibody concentrations to ExPEC10V and ExPEC4V O-serotypes on Days 1 (pre-vaccination), 15, 30, and 181. Additional blood samples will be collected from participants in the ExPEC10V selected dose group and the Prevnar 13 group in Cohort 1 at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826), and from all participants in Cohort 2 at Year 1 (Day 366). In Cohort 1, at the Day 30 visit, an additional blood sample of 50 mL will be drawn from a total of 30 participants willing to donate blood, for development or validation of the immunological assays (multiplex ECL-based immunoassay and MOPA) and for assessment of the validity and consistency of each assay experiment. This additional blood draw will be collected in a stepwise manner from participants, initially up to 15 participants from low and medium dose groups in Phase 1; followed by up to 15 participants from high dose group in Phase 1 and if needed, remainder from Phase 2a participants. Immunogenicity assessments of pneumococcal antibody titers elicited by Prevnar 13 will not be performed.

For all participants, serotype-specific total IgG antibody levels elicited by the vaccine against each of the 10 vaccine serotypes will be measured by multiplex ECL-based immunoassay and serotype-specific functional antibodies will be measured by MOPA (vaccine-induced functional antibodies against the O8 serotype will be measured using MOPA at baseline and Day 15 in Cohort 1, but O8 MOPA titers will not be reported for samples from all other time points in both cohorts). The multiplex ECL-based immunoassay and MOPA assessments will be performed by the sponsor and/or contract research organization.

Limits of assay variability will be defined within criteria described during assay qualification. Successful assay qualification will demonstrate designation of assays as fit for purpose. Operator training and adherence to defined Standard Operating Procedure and Good Clinical Practice (GCP) practices will be required to mitigate intra- and inter-operator assay variability. Factors to be evaluated will include: serum (antibody) freeze-thaw stability and inter-operator variability. Confounding factors associated with serum sample integrity and clinical sample handling, including sample identity and stability will be addressed by the laboratory manual specifying sample handling, shipment and storage procedures.

Samples collected for analyses of serum antibody concentrations to ExPEC10V O-serotypes may additionally be used to evaluate safety aspects that address concerns arising during or after the study period, for further characterization of immunogenicity. Genetic analyses will not be performed on these serum samples. Participant confidentiality will be maintained.

8.5. Other Evaluations

For Cohort 2, stool samples from a subset of 33% (approximately 140) randomly selected[a] participants will be analyzed using metagenomics to evaluate the effect of ExPEC10V on the prevalence of pathogens (eg, *Clostridium difficile*) and ExPEC10V serotypes O1A, O2, O4, O6A, O8, O15, O16, O18A, O25B, and O75 in the intestinal flora. Metagenomic analyses will be performed on stool samples collected on Days 1 (pre-vaccination), 30, and 181 for the ExPEC10V selected dose group and for the placebo group.

If a participant experiences an SAE related to IED, bacteremia, SIRS, or sepsis, and the pathogen is confirmed to be *E. coli*, an effort will be made to collect the isolate (blood [or any other sterile site] or urine) to perform O-serotyping. It is acknowledged that some study sites might not have the option to collect these isolates due to practical considerations.

Instructions for the collection, handling, storage, and shipment of stool, blood and urine samples are found in the laboratory manual that will be provided by the sponsor. Collection, handling, storage, and shipment of stool samples must be under the specified, and where applicable, controlled temperature conditions as indicated in the laboratory manual.

8.6. Pharmacogenomics

Pharmacogenomics are not evaluated in this study.

8.7. Medical Resource Utilization and Health Economics

Medical Resource Utilization and Health Economics parameters are not evaluated in this study.

9. STATISTICAL CONSIDERATIONS

Statistical analysis will be done by the sponsor or under the authority of the sponsor. A general description of the statistical methods to be used to analyze the efficacy and safety data is outlined below. Specific details will be provided in the Statistical Analysis Plan.

9.1. Statistical Hypotheses

No formal hypothesis testing will be done. One of the three ExPEC10V doses tested in Cohort 1 will be selected for further clinical development based on the safety (through Day 30) and immunogenicity data (Day 15 results obtained by multiplex ECL-based immunoassay and MOPA). The dose selection will consider the totality of the evidence available at the time of the primary analysis (Day 30), including an immunogenicity dose-selection algorithm that will guide the decision.

[a] Selected at randomization on Day 1 (using the IWRS); a baseline (pre-vaccination) stool sample will be collected from all participants and at the Day 30 and 181 visits from the selected subset of 33% of participants.

In Cohort 2, the safety and immunogenicity of the ExPEC10V dose selected from Cohort 1 will be further evaluated in participants in stable health with a history of UTI in the past 5 years.

9.2. Sample Size Determination

For Cohort 1, the primary objective of the study is to evaluate the safety and reactogenicity of different doses of ExPEC10V. The probability of observing at least 1 AE occurring at a rate of 1/100 is 63% with 100 participants receiving ExPEC10V per dose group. It was estimated that if no (S)AE will be observed in a dose group (N 100), this would provide 95% confidence that the true incidence is no more than 2.95%.

For Cohort 1, the co-primary objective of the study is to evaluate the dose-dependent immunogenicity of ExPEC10V, as measured by multiplex ECL-based immunoassay and MOPA. Based on the geometric mean ratios (GMR), in order to have 90% power to get a significant result (one-sided alpha 5%) if two groups are differing 2-fold (0.301 on $\log_{10}$-scale) for a certain O-serotype, assuming a standard deviation (SD) 0.7 (which was the case for O25B), 94 participants per ExPEC10V dose group will be needed. And with a 5% dropout rate, 100 participants per ExPEC10V dose group will be a reasonable sample size for Cohort 1 of the study.

For Cohort 2, the primary objective of the study is to evaluate the safety and reactogenicity of the selected dose of ExPEC10V. The probability of observing at least one AE occurring at a rate of 1/100 is 94% in Cohort 2 of the study with 280 participants receiving ExPEC10V. At a rate of 1/1,000, the probability of observing at least one AE is 24%. It was estimated that if no (S)AE will be observed in the safety cohort (Cohort 2) ExPEC10V group (N 280), this would provide 95% confidence that the true incidence is no more than 1.1%.

9.3. Populations for Analyses

Vaccination assignment will follow the as-treated principle. For purposes of analysis, the following populations are defined:

Full Analysis Set (FAS): The full analysis set will include all randomized participants with a vaccine administration documented.

Per-Protocol Immunogenicity Population (PPI): The per-protocol immunogenicity population will include all randomized and vaccinated participants, for whom immunogenicity data are available excluding the participants with major protocol deviations expecting to impact the immunogenicity outcomes.

9.4. Statistical Analyses

9.4.1. Safety Analyses

No formal statistical testing of safety data is planned. The safety analysis will include the descriptive summary (including 95% confidence intervals) of solicited local AEs, solicited systemic AEs, unsolicited AEs, and SAEs. The overall frequencies per study vaccination group as

well as frequencies according to severity and duration will be calculated for solicited and unsolicited AEs.

Data from Cohort 1 and Cohort 2 of the study will be analyzed separately.

Adverse Events

The verbatim terms used in the eCRF by investigators to identify AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). All reported AEs and events-related diary information (solicited local at injection site and systemic) with onset within 30 days after vaccination will be included in the analysis. For each AE, the percentage of participants who experience at least 1 occurrence of the given event will be summarized by study vaccination group. Concomitant medications will be coded, using the World Health Organization Drug Dictionary (WHO-DD).

Solicited local (at injection site) and systemic AEs will be summarized descriptively. The overall frequencies per study vaccination group as well as frequencies according to severity and duration will be calculated for solicited AEs. In addition, the number and percentages of participants with at least one solicited local (at injection site) or systemic AE will be presented. Frequencies of unsolicited AEs, separately for all and vaccine-related only, will be presented by system organ class and preferred term, while those of solicited AEs will be presented only by preferred term.

Summaries, listings, datasets, or participant narratives may be provided, as appropriate, for those participants who die, who discontinue study vaccination due to an AE, or who experience a severe AE or an SAE.

Clinical Laboratory Tests (Cohort 1)

For Cohort 1, laboratory data will be summarized by type of laboratory test. Reference ranges and abnormal results will be used in the summary of laboratory data. A listing of participants with any laboratory results outside the reference ranges will be provided.

Vital Signs

Descriptive statistics of temperature, pulse/heart rate, respiratory rate, and blood pressure (systolic and diastolic) (supine) values and changes from baseline will be summarized at each scheduled time point.

9.4.2. Immunogenicity Analyses

For ExPEC10V serotype antibodies as measured by multiplex ECL-based immunoassay and MOPA, and EPA as measured by multiplex ECL-based immunoassay only, the following measures of immunogenicity will be evaluated and tabulated by the study vaccination groups, for all immunogenicity time points:

- proportion of participants with a ≥2-fold and ≥4-fold increase in serum antibody titers from Day 1 (pre-vaccination)

- geometric mean titer (GMT)
- GMR: fold change from baseline, calculated from the post-baseline/baseline value.

For the LTFU period, descriptive summaries of immunogenicity will be presented for each serotype.

Immunogenicity Dose-Selection Algorithm (Cohort 1)

The dose selection will consider the totality of the evidence available at the time of the primary analysis for Cohort 1, including an immunogenicity dose-selection algorithm that will guide the decision.

The immunogenicity dose-selection algorithm will include the $\log_{10}$ transformation of the fold increase from baseline to Day 15 as the response variable and the independent variables will include the dose groups and the $\log_{10}$ transformations of the baseline titer.

The algorithm is a stepwise procedure where in the first four steps, the $\log_{10}$ transformation of the fold increase from baseline to Day 15 for the serotypes O25B, O6A, O2, and O1A (in that order) are included in the model as a response variable. If after these four steps, more than one group is retained (see below), the other six serotypes are included in the model as a response variable and the dose group that is selected the most will be used as the selected dose group. In each step, both the least squares mean of each of the dose groups included in the model and a 90% confidence interval of the difference between the least squares mean of a dose group and that dose group having the highest least squares mean are computed. The dose group having the highest least squares mean and those other dose groups that are non-inferior to this dose group based on the confidence limits are retained. Only these dose groups are included in the model as covariate in the next step. The dose selected by the immunogenicity dose-selection algorithm will be the dose group retained after the final step. More details are provided in the Statistical Analysis Plan.

9.5. Planned Analyses

The Statistical Analysis Plan will describe the planned analyses in greater detail.

9.5.1. Cohort 1

The primary analysis for Cohort 1 will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. These analyses will include immunogenicity data up to Day 15 and all available safety data.

The final analysis for Cohort 1 will occur when all participants have completed the Day 181 visit or have discontinued earlier.

At the time of the primary analysis, Cohort 1 will be unblinded for specific sponsor personnel. Participants, clinical staff and study-site personnel will remain blinded to the study vaccination allocation until the final analysis database lock.

For participants in Cohort 1 progressing to the open-label LTFU period, yearly follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to the time of the visit at Year 1 (Day 366), Year 2 (Day 731), Year 3 (Day 1096), Year 4 (Day 1461), and Year 5 (Day 1826) after vaccination.

9.5.2. Cohort 2

The primary analysis for Cohort 2 will occur when all participants have completed the Day 30 visit (Visit 4) or have discontinued earlier. These analyses will include immunogenicity data up to Day 30 and all available safety data. The rationale for including immunogenicity data up to Day 30 in the primary analysis for Cohort 2 (compared to Cohort 1 where immunogenicity data up to Day 15 will be included) is to provide an additional time point for analysis of immunogenicity and kinetics with the selected ExPEC10V dose.

The final analysis for Cohort 2 will occur when all participants have completed the Day 181 visit or have discontinued earlier.

At the time of the primary analysis, Cohort 2 will be unblinded for specific sponsor personnel. Participants, clinical staff, and study-site personnel will remain blinded to the study vaccination allocation until all participants have completed the double-blind LTFU period.

For all participants in Cohort 2, follow-up analyses will include safety and immunogenicity data (multiplex ECL-based immunoassay and MOPA) collected up to the time of the visit at Year 1 (Day 366) after vaccination.

The effect of age and BMI on immunogenicity will also be assessed; further details will be provided in the Statistical Analysis Plan.

9.6. Data Review Committee

A DRC will be established as noted in Committees Structure in Section 10.2 Appendix 2, Regulatory, Ethical, and Study Oversight Considerations.

9.7. Study Vaccination Pausing Rules

The PI(s), the SRP and the SML will monitor the study vaccination pausing rules. If study vaccination is considered to raise significant safety concerns, further vaccination of participants will be paused. The concerned data will be reviewed by the DRC, after which the DRC will recommend whether the pause can be lifted or not, or whether other steps are needed.

The occurrence of any of the following events will lead to pause in further study vaccination. The list is only applicable for concerned AEs that occur up to 4 weeks after the vaccination and to concerned SAEs:

1. Death of a participant, considered related to any study vaccine or if the causal relationship to the study vaccine cannot be excluded; *OR*

   Note: All cases of death will be sent for DRC information. Upon their review, the DRC may decide whether a study pause is required.
2. One or more participants experience a SAE or a Grade 4 (solicited or unsolicited) AE or, a persistent (upon repeat testing) Grade 4 laboratory abnormality that is determined to be related to study vaccine[a]; *OR*
3. One or more participants experience anaphylaxis or generalized urticaria within 24 hours of vaccination, clearly not attributable to other causes than vaccination with study vaccine; *OR*
4. Three or more participants experience a Grade 3 unsolicited AE of the same type (as per medical judgment of the sponsor), that is determined to be related to study vaccine[b], that persists for 72 hours or longer; *OR*
5. For Cohort 1 only, three or more participants experience a persistent (upon repeat testing) Grade 3 laboratory abnormality related to the same laboratory parameter and considered related to study vaccine; *OR*
6. Three or more participants experience a Grade 3 solicited systemic AE of the same type, determined to be related to study vaccine[j]; and persisting as Grade 3 for longer than three consecutive days[c].

[a] excluding related to the Prevnar 13 vaccine (active control group)

[b] excluding related to the Prevnar 13 vaccine (active control group)

[c] The day of occurrence of the AE is counted as Day 1.

10. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1. Appendix 1: Abbreviations

ACIP	Advisory Committee for Immunization Practices
AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BMI	body mass index
BUN	blood urea nitrogen
COVID-19	Coronavirus Disease 2019
CPRD	Clinical Practice Research Datalink
CRP	C-reactive protein
CSR	Clinical Study Report
DRC	Data Review Committee
EC_{50}	half maximal effective concentration
ECG	electrocardiogram
ECL	electrochemiluminescent
eCRF	electronic case report form
eDC	electronic data capture
ediary	electronic diary
ELISA	enzyme-linked immunosorbent assay
EPA	exotoxin protein A
ExPEC	extraintestinal pathogenic *Escherichia coli*
ExPEC4V	4-valent ExPEC vaccine
ExPEC10V	10-valent ExPEC vaccine (candidate vaccine)
FAS	Full Analysis Set
FDA	Food and Drug Administration
FSH	follicle-stimulating hormone
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GMR	geometric mean ratio
GMT	geometric mean titer
HCV	hepatitis C virus
HHS	Health and Human Services
HIV	human immunodeficiency virus
HRT	hormonal replacement therapy
ICF	informed consent form
ICH	International Conference on Harmonisation
ICMJE	International Committee of Medical Journal Editors
IED	invasive ExPEC disease
IEC	Independent Ethics Committee
IgG	immunoglobulin G
IM	intramuscular
IRB	Institutional Review Board
IWRS	interactive web response system
KPNW	Kaiser Permanente Northwest
LTFU	long-term follow-up
MedDRA	Medical Dictionary for Regulatory Activities
MOPA	multiplex opsonophagocytic assay
NZW	New Zealand White
OHRP	Office for Human Research Protections
OPA	opsonophagocytic killing assay
PCV	pneumococcal vaccines
PI	principal investigator
PPI	Per-Protocol Immunogenicity

PPSV23	PNEUMOVAX® 23
PQC	product quality complaint
PS	polysaccharide
SAE	serious adverse event
SD	standard deviation
SIRS	systemic inflammatory response syndrome
SML	sponsor medical lead
SRP	study responsible physician
SUSAR	suspected unexpected serious adverse reaction
US	United States
UTI	urinary tract infection
WBC	white blood cells
WHO-DD	World Health Organization Drug Dictionary

10.2. Appendix 2: Regulatory, Ethical, and Study Oversight Considerations

REGULATORY AND ETHICAL CONSIDERATIONS

Investigator Responsibilities

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

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

Protocol Clarification Communications

If text within a final approved protocol requires clarification (eg, current wording is unclear or ambiguous) that does not change any aspect of the current study conduct, a protocol clarification communication (PCC) may be prepared. The PCC Document will be communicated to the Investigational Site, Site Monitors, Local Trial Managers, Clinical Trial Managers, and/or Contract Research Organizations who will ensure that the PCC explanations are followed by the investigators.

The PCC Document may be shared by the sites with IECs/IRBs per local regulations.

The PCC Documents must NOT be used in place of protocol amendments, but the content of the PCC Document must be included in any future protocol amendments.

Protocol Amendments

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

During the course of the study, in situations where a departure from the protocol is unavoidable, the investigator or other physician in attendance will contact the appropriate sponsor representative listed in the Contact Information page(s), which will be provided as a separate document. Except in emergency situations, this contact should be made before implementing any departure from the protocol. In all cases, contact with the sponsor must be made as soon as possible to discuss the

situation and agree on an appropriate course of action. The data recorded in the eCRF and source documents will reflect any departure from the protocol, and the source documents will describe this departure and the circumstances requiring it.

Regulatory Approval/Notification

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

Required Prestudy Documentation

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

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

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

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

- Local laboratory documentation demonstrating competence and test reliability (eg, accreditation/license), if applicable

Independent Ethics Committee or Institutional Review Board

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

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

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

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

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

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

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

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

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

Country Selection

This study will only be conducted in those countries where the intent is to launch or otherwise help ensure access to the developed product if the need for the product persists, unless explicitly addressed as a specific ethical consideration in Section 4.2.3, Study-Specific Ethical Design Considerations.

Other Ethical Considerations

For study-specific ethical design considerations, refer to Section 4.2.3.

FINANCIAL DISCLOSURE

Investigators and subinvestigators will provide the sponsor with sufficient, accurate financial information in accordance with local regulations to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

Refer to Required Prestudy Documentation (above) for details on financial disclosure.

INFORMED CONSENT PROCESS

Each participant must give written consent according to local requirements after the nature of the study has been fully explained. The ICF(s) must be signed before performance of any study-related activity. The ICF(s) that is/are used must be approved by both the sponsor and by the reviewing IEC/IRB and be in a language that the participant can read and understand. The informed consent

should be in accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and sponsor policy.

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

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

Informed consent will also be obtained from the 30 participants willing to donate blood (50 mL) for development or validation of the immunological assays (multiplex ECL-based immunoassay and MOPA) and for assessment of the validity and consistency of each assay experiment.

In Cohort 1, additional informed consent for participants progressing to the open-label LTFU period (participants in the ExPEC10V selected dose group based on the primary analysis and participants in the Prevnar 13 group) will be required. Information relevant to the participant’s willingness to progress to the follow-up period of the study will be provided to the participant in a timely manner and written informed consent for participation in this period will be obtained.

DATA PROTECTION

Privacy of Personal Data

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

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

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

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

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

LONG-TERM RETENTION OF SAMPLES FOR ADDITIONAL FUTURE RESEARCH

Samples collected in this study may be stored for up to 15 years (or according to local regulations) for additional research. Samples will only be used to understand ExPEC10V, to understand IED, to understand differential vaccine responders, and to develop tests/assays related to ExPEC10V and IED. The research may begin at any time during the study or the post-study storage period.

Stored samples will be coded throughout the sample storage and analysis process and will not be labeled with personal identifiers. Participants may withdraw their consent for their samples to be stored for research (refer to Section 7.2.1, Withdrawal From the Use of Research Samples).

COMMITTEES STRUCTURE

Data Review Committee (DRC)

An internal DRC will be established for this study, which will monitor data to ensure the safety and well-being of the participants enrolled. The DRC will consist of sponsor personnel not directly involved in the conduct of the study and who have expertise in clinical study conduct and vaccines, at least one medical expert in the relevant therapeutic area, at least one statistician, and a safety expert. The DRC will convene, according to their charter, to discuss any safety issues and any situation meeting a specific study vaccination pausing rule (Section 9.7 Study Vaccination Pausing Rules). The PI(s) and SRP(s) will inform the DRC of any AE of concern.

For Cohort 1, the DRC will specifically review safety data (including solicited and unsolicited AEs, SAEs, and available laboratory assessments) from Phase 1 participants after vaccination of 28 participants at each dose level and monitor data on an ongoing basis in order to ensure the continuing safety of all participants enrolled in this study.

For Cohort 2, if a specific study vaccination pausing rule has been met or if any other safety issue may arise, the DRC will be convened to evaluate safety data.

After these reviews, the DRC will make recommendations regarding the continuation of the study. The conclusions of the DRC will be communicated to the investigators, the IRB/IEC, and the national regulatory authorities, as appropriate.

The DRC will review blinded data first but may also review unblinded data if deemed necessary.

PUBLICATION POLICY/DISSEMINATION OF CLINICAL STUDY DATA

All information, including but not limited to information regarding ExPEC10V and ExPEC4V or the sponsor's operations (eg, patent application, formulas, manufacturing processes, basic scientific data, prior clinical data, formulation information) supplied by the sponsor to the investigator and not previously published, and any data, including exploratory research data, generated as a result of this study, are considered confidential and remain the sole property of the sponsor. The investigator agrees to maintain this information in confidence and use this information only to accomplish this study and will not use it for other purposes without the sponsor's prior written consent.

The investigator understands that the information developed in the study will be used by the sponsor in connection with the continued development of ExPEC10V, and thus may be disclosed as required to other clinical investigators or regulatory agencies. To permit the information derived from the clinical studies to be used, the investigator is obligated to provide the sponsor with all data obtained in the study.

The results of the study will be reported in a Clinical Study Report generated by the sponsor and will contain data from all study sites that participated in the study as per protocol. Recruitment performance or specific expertise related to the nature and the key assessment parameters of the study will be used to determine a coordinating investigator for the study. Results of exploratory analyses performed after the Clinical Study Report has been issued will be reported in a separate report and will not require a revision of the Clinical Study Report.

Study participant identifiers will not be used in publication of results. Any work created in connection with performance of the study and contained in the data that can benefit from copyright protection (except any publication by the investigator as provided for below) shall be the property of the sponsor as author and owner of copyright in such work.

Consistent with Good Publication Practices and International Committee of Medical Journal Editors (ICMJE) guidelines, the sponsor shall have the right to publish such primary (multicenter) data and information without approval from the investigator. The investigator has the right to publish study site-specific data after the primary data are published. If an investigator wishes to publish information from the study, a copy of the manuscript must be provided to the sponsor for review at least 60 days before submission for publication or presentation. Expedited reviews will be arranged for abstracts, poster presentations, or other materials. If requested by the sponsor in

writing, the investigator will withhold such publication for up to an additional 60 days to allow for filing of a patent application. In the event that issues arise regarding scientific integrity or regulatory compliance, the sponsor will review these issues with the investigator. The sponsor will not mandate modifications to scientific content and does not have the right to suppress information.

For multicenter study designs and sub-study approaches, secondary results generally should not be published before the primary endpoints of a study have been published. Similarly, investigators will recognize the integrity of a multicenter study by not submitting for publication data derived from the individual study site until the combined results from the completed study have been submitted for publication, within 18 months after the study end date, or the sponsor confirms there will be no multicenter study publication. Authorship of publications resulting from this study will be based on the guidelines on authorship, such as those described in the ICMJE Recommendations for the Conduct, Reporting, Editing and Publication of Scholarly Work in Medical Journals, which state that the named authors must have made a significant contribution to the conception or design of the work; or the acquisition, analysis, or interpretation of the data for the work; and drafted the work or revised it critically for important intellectual content; and given final approval of the version to be published; and agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Registration of Clinical Studies and Disclosure of Results

The sponsor will register and disclose the existence of and the results of clinical studies as required by law. The disclosure of the final study results will be performed after the end of study in order to ensure the statistical analyses are relevant.

DATA QUALITY ASSURANCE

Data Quality Assurance/Quality Control

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

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

CASE REPORT FORM COMPLETION

Electronic case report forms (eCRFs) are prepared and provided by the sponsor for each participant in electronic format. All data relating to the study must be recorded in eCRF. All eCRF entries,

corrections, and alterations must be made by the investigator or authorized study-site personnel. The investigator must verify that all data entries in the eCRF are accurate and correct.

The study data will be transcribed by study-site personnel from the source documents onto an electronic eCRF, if applicable. Study-specific data will be transmitted in a secure manner to the sponsor.

Data must be entered into eCRF in English. The eCRF must be completed as soon as possible after a participant visit and the forms should be available for review at the next scheduled monitoring visit.

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

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

SOURCE DOCUMENTS

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

The author of an entry in the source documents should be identifiable.

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

MONITORING

The sponsor or designee will use a combination of monitoring techniques central, remote, or on-site monitoring to monitor this study.

The sponsor will perform on-site monitoring visits as frequently as necessary. The monitor will record dates of the visits in a study site visit log that will be kept at the study site. The first post-initiation visit will be made as soon as possible after enrollment has begun. At these visits, the monitor will compare the data entered into the eCRF with the source documents (eg, hospital/clinic/physician's office medical records). The nature and location of all source

documents will be identified to ensure that all sources of original data required to complete the eCRF are known to the sponsor and study-site personnel and are accessible for verification by the sponsor study-site contact. If electronic records are maintained at the study site, the method of verification must be discussed with the study-site personnel.

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

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

ON-SITE AUDITS

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

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

RECORD RETENTION

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

Essential documents must be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational product. These documents will be retained for a longer period

if required by the applicable regulatory requirements or by an agreement with the sponsor. It is the responsibility of the sponsor to inform the investigator/institution as to when these documents no longer need to be retained.

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

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

STUDY AND SITE START AND CLOSURE

First Act of Recruitment

The first site open is considered the first act of recruitment and it becomes the study start date.

Study Termination

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

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

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

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

10.3. Appendix 3: Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

ADVERSE EVENT DEFINITIONS AND CLASSIFICATIONS

Adverse Event (AE)

An AE is any untoward medical occurrence in a clinical study participant administered a medicinal (investigational or non-investigational) product. An AE does not necessarily have a causal relationship with the study vaccine. An AE can therefore be any unfavorable and unintended sign (including an abnormal finding), symptom, or disease temporally associated with the use of a medicinal (investigational or non-investigational) product, whether or not related to that medicinal (investigational or non-investigational) product (Definition per ICH).

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

Note: For the time of sponsor's AEs collection, see All Adverse Events under Section 8.2.1, Time Period and Frequency for Collecting Adverse Events and Serious Adverse Events Information.

Serious Adverse Event (SAE)

An SAE based on ICH and EU Guidelines on Pharmacovigilance for Medicinal Products for Human Use is any untoward medical occurrence that at any dose:

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

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

If a serious and unexpected AE occurs for which there is evidence suggesting a causal relationship between the study vaccine and the event (eg, death from anaphylaxis), the event must be reported as a serious and unexpected suspected adverse reaction.

Unlisted (Unexpected) Adverse Event/Reference Safety Information

An AE is considered unlisted if the nature or severity is not consistent with the applicable product reference safety information. For ExPEC10V and ExPEC4V, the expectedness of an AE will be determined by whether or not it is listed in the respective Investigator's Brochure. For Prevnar 13 with a marketing authorization, the expectedness of an AE will be determined by whether or not it is listed in the package insert.

ATTRIBUTION DEFINITIONS

Assessment of Causality

The causal relationship to study vaccine is determined by the investigator. The following selection should be used to assess all AEs:

Related: There is a reasonable causal relationship between study vaccine administration and the AE.

Not related: There is not a reasonable causal relationship between study vaccine administration and the AE.

The term "reasonable causal relationship" means there is evidence to support a causal relationship.

By definition, all solicited AEs at the injection site (local) will be considered related to the study vaccine administration.

SEVERITY CRITERIA

All AEs and laboratory data will be coded for severity using a modified version of the FDA grading table, based on the version of September 2007[4], included in Section 10.5 Appendix 5 Toxicity Tables.

For AEs not identified in the grading table, the following guidelines will be applied:

Grade 1 - Mild: Symptoms causing no or minimal interference with usual social and functional activities.

Grade 2 - Moderate: Symptoms causing greater than minimal interference with usual social and functional activities.

Grade 3 - Severe: Symptoms causing inability to perform usual social and functional activities and requires medical intervention.

Grade 4 - Potentially life-threatening: Symptoms causing inability to perform basic self-care functions, OR medical or operative intervention indicated to prevent permanent impairment, persistent disability, OR emergency room visit or hospitalization.

The investigator should use clinical judgment in assessing the severity of events not directly experienced by the participant (eg, laboratory abnormalities).

The severity of solicited AEs will be graded in the ediary by the participant based on the severity assessment provided in the ediary and then verified by the investigator using the scoring system shown in Section 10.5 Appendix 5 Toxicity Tables. (Note: severity of the measured events - erythema, and swelling - will be derived from the diameter).

SPECIAL REPORTING SITUATIONS

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

- Suspected abuse/misuse of a sponsor study vaccine
- Accidental or occupational exposure to a sponsor study vaccine
- Medication error, intercepted medication error, or potential medication error involving a Johnson & Johnson medicinal product (with or without patient exposure to the Johnson & Johnson medicinal product, eg, product name confusion, product label confusion, intercepted prescribing or dispensing errors)

Special reporting situations should be recorded in the eCRF. Any special reporting situation that meets the criteria of a SAE should be recorded on the SAE page of the eCRF.

PROCEDURES

All Adverse Events

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

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

- Study number
- Statement, in the local language(s), that the participant is participating in a clinical study

- Investigator's name and 24-hour contact telephone number
- Local sponsor's name and 24-hour contact telephone number (for medical personnel only)
- Site number
- Participant number
- Any other information that is required to do an emergency breaking of the blind

Serious Adverse Events

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

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

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

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

The cause of death of a participant in a study, whether the event is expected or associated with the study vaccine or not, is considered an SAE.

CONTACTING SPONSOR REGARDING SAFETY

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

PRODUCT QUALITY COMPLAINT HANDLING

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

Procedures

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

If the defect is combined with an SAE, the study-site personnel must report the PQC to the sponsor according to the SAE reporting timelines (refer to Section 8.2.1, Time Period and Frequency for Collecting Adverse Event and Serious Adverse Event Information). A sample of the suspected product should be maintained for further investigation if requested by the sponsor.

Contacting Sponsor Regarding Product Quality

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

10.4. Appendix 4: Contraceptive and Barrier Guidance and Collection of Pregnancy Information

Participants must follow contraceptive measures as outlined in Section 5.1, Inclusion Criteria. Pregnancy information will be collected and reported as noted in Section 8.2.5, Pregnancy and Section 10.3 Appendix 3 Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting.

Definitions

Woman Not of Childbearing Potential

- **postmenopausal**
  A postmenopausal state is defined as no menses for 12 months without an alternative medical cause. A high follicle-stimulating hormone (FSH) level (>40 IU/L or mIU/mL) in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT).
- **permanently sterile**
  Permanent sterilization methods include hysterectomy, bilateral salpingectomy, bilateral tubal occlusion/ligation procedures, and bilateral oophorectomy.

10.5. Appendix 5: Toxicity Tables

Adapted from the FDA Guidance document "Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials" (September 2007)

A: Tables for Clinical Abnormalities

Local Reaction to Injectable Product	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4)
Pain/Tenderness	Aware of symptoms but easily tolerated; Does not interfere with activity; Discomfort only to touch	Notable symptoms; Requires modification in activity or use of medications; Discomfort with movement;	Incapacitating symptoms; Use of narcotic pain reliever; Inability to do work, school, or usual activities; Use of narcotic pain reliever	Hospitalization; Pain/tenderness causing inability to perform basic self-care function;
Erythema	2.5 – 5 cm	5.1 – 10 cm	> 10 cm	Hospitalization; Necrosis or exfoliative dermatitis
Swelling	2.5 – 5 cm	5.1 – 10 cm	> 10 cm	Hospitalization; Necrosis

Vital Signs *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4)
Fever (°C) ** (°F) **	38.0 – 38.4 100.4 – 101.1	38.5 – 38.9 101.2 – 102.0	39.0 – 40.0 102.1 – 104.0	>40.0 >104.0
Tachycardia - beats per minute	101 – 115	116 – 130	> 130	Hospitalization for arrhythmia
Bradycardia - beats per minute***	50 – 54	45 – 49	< 45	Hospitalization for arrhythmia
Hypertension (systolic) - mm Hg	141 – 150	151 – 160****	> 160****	Hospitalization for malignant hypertension
Hypertension (diastolic) - mm Hg	91 – 95	96 – 100	> 100	Hospitalization for malignant hypertension
Hypotension (systolic) - mm Hg	85 – 89	80 – 84	< 80	Hospitalization for hypotensive shock
Respiratory Rate – breaths per minute	17 – 20	21 – 25	> 25	Intubation

* Participant should be at rest for all vital sign measurements.

** Non axillary temperatures only; For oral temperature; no recent hot or cold beverages or smoking.

*** When resting heart rate is between 60 100 beats per minute. Use clinical judgment when characterizing bradycardia among some healthy participant populations, for example, conditioned athletes.

**** Modified upper limit regarding the participant population (participants >60 years)

Systemic (General)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4)
Vomiting	No interference with activity or 1 - 2 episodes/ 24 hours	Some interference with activity or > 2 episodes/ 24 hours	Prevents daily activity, requires outpatient IV hydration	Hospitalization for hypotensive shock
Nausea	Minimal symptoms; causes minimal or no interference with work, school, or self-care activities	Notable symptoms; Requires modification in activity or use of medications; Does NOT result in loss of work, school, or cancellation of social activities.	Incapacitating symptoms; Requires bed rest and/or results in loss of work, school, or cancellation of social activities.	Hospitalization; Inability to perform basic self-care functions.
Diarrhea	2 – 3 loose stools or < 400 gms/ 24 hours	4 – 5 stools or 400 – 800 gms/ 24 hours	6 or more watery stools or > 800gms/24 hours or oral rehydration necessary	Hospitalization; Hypotensive shock OR IV fluid replacement indicated
Headache	Minimal symptoms; causes minimal or no interference with work, school, or self-care activities.	Notable symptoms; Requires modification in activity or use of medications; Does NOT result in loss of work, school, or cancellation of social activities.	Incapacitating symptoms; Requires bed rest and/or results in loss of work, school, or cancellation of social activities; Use of narcotic pain reliever.	Hospitalization; Inability to perform basic self-care functions.
Fatigue	Minimal symptoms; causes minimal or no interference with work, school, or self-care activities.	Notable symptoms; Requires modification in activity or use of medications; Does NOT result in loss of work, school, or cancellation of social activities.	Incapacitating symptoms; Requires bed rest and/or results in loss of work, school, or cancellation of social activities. Use of narcotic pain reliever.	Hospitalization; Inability to perform basic self-care functions.

Systemic (General)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4)
Myalgia	Minimal symptoms; causes minimal or no interference with work, school, or self-care activities.	Notable symptoms; Requires modification in activity or use of medications; Does NOT result in loss of work, school, or cancellation of social activities.	Incapacitating symptoms; Requires bed rest and/or results in loss of work, school, or cancellation of social activities; Use of narcotic pain reliever.	Hospitalization; Inability to perform basic self-care functions.

Systemic Illness	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4)
Illness or clinical adverse event (as defined according to applicable regulations)	No interference with activity	Some interference with activity not requiring medical intervention	Prevents daily activity and requires medical intervention	Hospitalization

B: Tables for Laboratory Abnormalities

The grading scale used for laboratory assessments is based on the FDA Guidance document "Toxicity Grading Scale from Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials". Any laboratory value shown as a "graded" value in the table that is within the central laboratory normal ranges will not be graded for severity or recorded as an AE. For hemoglobin, both the actual value and the change from reference will be graded. For the change from reference, the corresponding actual value should also be at least Grade 1.

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4) **
Sodium – Hyponatremia mEq/L	132 – 134	130 – 131	125 – 129	< 125
Sodium – Hypernatremia mEq/L	144 – 145	146 – 147	148 – 150	> 150
Potassium – Hyperkalemia mEq/L	5.1 – 5.2	5.3 – 5.4	5.5 – 5.6	> 5.6
Potassium – Hypokalemia mEq/L	3.5 – 3.6	3.3 – 3.4	3.1 – 3.2	< 3.1
Glucose – Hypoglycemia mg/dL	65 – 69	55 – 64	45 – 54	< 45
Glucose – Hyperglycemia Fasting mg/dL Random – mg/dL	100 – 110 110 – 125	111 – 125 126 – 200	>125 >200	Insulin requirements or hyperosmolar coma
Blood Urea Nitrogen BUN mg/dL	23 – 26	27 – 31	> 31	Requires dialysis

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4) **
Creatinine – mg/dL	1.5 – 1.7	1.8 – 2.0	2.1 – 2.5	> 2.5 or requires dialysis
Calcium – hypocalcemia mg/dL	8.0 – 8.4	7.5 – 7.9	7.0 – 7.4	< 7.0
Calcium – hypercalcemia mg/dL	10.5 – 11.0	11.1 – 11.5	11.6 – 12.0	> 12.0
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	0.9 – 1.0	< 0.9
Phosphorous – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	1.6 – 1.9	< 1.6
CPK – mg/dL	1.25 – 1.5 x ULN***	1.6 – 3.0 x ULN	3.1 –10 x ULN	> 10 x ULN
Albumin – Hypoalbuminemia g/dL	2.8 – 3.1	2.5 – 2.7	< 2.5	--
Total Protein – Hypoproteinemia g/dL	5.5 – 6.0	5.0 – 5.4	< 5.0	--
Alkaline phosphate – increase by factor	1.1 – 2.0 x ULN	2.1 – 3.0 x ULN	3.1 – 10 x ULN	> 10 x ULN
Liver Function Tests – ALT, AST increase by factor	1.1 – 2.5 x ULN	2.6 – 5.0 x ULN	5.1 – 10 x ULN	> 10 x ULN
Bilirubin – when accompanied by any increase in Liver Function Test increase by factor	1.1 – 1.25 x ULN	1.26 – 1.5 x ULN	1.51 – 1.75 x ULN	> 1.75 x ULN
Bilirubin – when Liver Function Test is normal; increase by factor	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.0 – 3.0 x ULN	> 3.0 x ULN
Cholesterol	201 – 210	211 – 225	> 226	--
Pancreatic enzymes – amylase, lipase	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.1 – 5.0 x ULN	> 5.0 x ULN

* The laboratory values provided in the tables serve as guidelines and are dependent upon central laboratory normal parameters. Central laboratory normal reference ranges should be provided to demonstrate that they are appropriate.

** The clinical signs or symptoms associated with laboratory abnormalities might result in characterization of the laboratory abnormalities as potentially life threatening (Grade 4). For example. a low sodium value that falls within a Grade 3 parameter (125 129 mE/L) should be recorded as a Grade 4 hyponatremia event if the participant had a new seizure associated with the low sodium value.

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

Hematology *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-threatening (Grade 4) **
Hemoglobin (Female) - gm/dL	11.0 – 12.0	9.5 – 10.9	8.0 – 9.4	< 8.0
Hemoglobin (Female) change from baseline value - gm/dL	Any decrease – 1.5	1.6 – 2.0	2.1 – 5.0	> 5.0
Hemoglobin (Male) - gm/dL	12.5 – 13.5	10.5 – 12.4	8.5 – 10.4	< 8.5
Hemoglobin (Male) change from baseline value – gm/dL	Any decrease – 1.5	1.6 – 2.0	2.1 – 5.0	> 5.0
WBC Increase - cell/mm^3	10,800 – 15,000	15,001 – 20,000	20,001 – 25, 000	> 25,000
WBC Decrease - cell/mm^3	2,500 – 3,500	1,500 – 2,499	1,000 – 1,499	< 1,000
Lymphocytes Decrease - cell/mm^3	750 – 1,000	500 – 749	250 – 499	< 250
Neutrophils Decrease - cell/mm^3	1,500 – 2,000	1,000 – 1,499	500 – 999	< 500
Eosinophils - cell/mm^3	650 – 1500	1501 - 5000	> 5000	Hypereosinophilic
Platelets Decreased - cell/mm^3	125,000 – 140,000	100,000 – 124,000	25,000 – 99,000	< 25,000
PT – increase by factor (prothrombin time)	1.0 – 1.10 x ULN**	1.11 – 1.20 x ULN	1.21 – 1.25 x ULN	> 1.25 ULN
PTT – increase by factor (partial thromboplastin time)	1.0 – 1.2 x ULN	1.21 – 1.4 x ULN	1.41 – 1.5 x ULN	> 1.5 x ULN
Fibrinogen increase - mg/dL	400 – 500	501 – 600	> 600	--
Fibrinogen decrease - mg/dL	150 – 200	125 – 149	100 – 124	< 100 or associated with gross bleeding or disseminated intravascular coagulation (DIC)

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

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

10.6. Appendix 6: COVID-19 Appendix

GUIDANCE ON STUDY CONDUCT DURING THE COVID-19 PANDEMIC

It is recognized that the Coronavirus Disease 2019 (COVID-19) pandemic may have an impact on the conduct of this clinical study due to, for example, self-isolation/quarantine by participants and study-site personnel; travel restrictions/limited access to public places, including hospitals; study site personnel being reassigned to critical tasks.

In alignment with recent health authority guidance, the sponsor is providing options for study related participant management in the event of disruption to the conduct of the study. This guidance does not supersede any local or government requirements or the clinical judgement of the investigator to protect the health and well-being of participants and site staff.

Scheduled visits that cannot be conducted in person at the study site will be performed to the extent possible remotely/virtually (ie via telephone visits or home-based visits, if the participant allows) or delayed until such time that on-site visits can be resumed. The actual visit date and the type of visit (ie, telephone or home-based visit) should be captured in the eCRF. Procedures that require an on-site visit should be excluded. The missed procedures should be recorded as "missed due to COVID-19".

At each contact, participants will be interviewed to collect safety data. Participants will also be questioned regarding general health status to fulfill any physical examination requirement.

Every effort should be made to adhere to protocol-specified assessments, including follow up. Modifications to protocol-required assessments may be permitted via COVID-19 Appendix after consultation with the participant, investigator, and the sponsor. Missed assessments/visits will be captured in the clinical trial management system for protocol deviations. Withdrawal from the study should be documented with the prefix "COVID-19-related" in the case report form (CRF).

The sponsor will continue to monitor the conduct and progress of the clinical study, and any COVID-19 related changes will be communicated to the sites and to the health authorities according to local guidance. If a participant has tested positive for COVID 19, the investigator should contact the sponsor's responsible medical officer to discuss plans for follow-up. Hospitalization of a participant due to COVID-19 should be reported as a serious adverse event. A positive laboratory test result for SARS-CoV2 without symptoms or COVID-19 without hospitalization should be reported in line with the safety reporting requirements described in the study protocol. Modifications made to the study conduct as a result of the COVID-19 pandemic should be summarized in the clinical study report.

Screening and Vaccination Visit:

- The screening and vaccination visit should be conducted on-site.

Study Visits and Assessments:

Study visits may be performed by a home health nurse until such time that on-site visits can be resumed. The following activities may be completed by a home health nurse as required per the Schedule of Activities:

- Collecting safety information: adverse events (solicited and unsolicited), serious adverse events, physical examination, vital signs, and concomitant medication.
- Collecting stool and immunogenicity blood samples.

The collected data related to adverse events, serious adverse events, physical examination, and vital signs will be reviewed and assessed by the investigator. Procedures and timings should follow the Schedule of Activities as closely as possible. Standard Adverse Event/Serious Adverse Event reporting requirements apply.

Informed Consent:

- A revised ICF or an addendum to the ICF, arising from this amendment, is to be signed by the participants during a site visit. For any informed consent that cannot be performed in person (eg, verbal consent by telephone), the process must be documented and confirmed by way of normal consent procedures at the earliest opportunity. If consent is given verbally by phone, an impartial witness should be present, if required by local regulations, for the entire informed consent process (which includes reading and explaining all written information) and should personally date and sign the ICF after the oral consent is obtained.

Source Data Verification/Monitoring:

- In case on-site monitoring visits are not possible, the site monitor may contact the investigator to arrange monitoring visits and activities remotely. Additional on-site monitoring visits may be needed in the future to catch up on source data verification.

10.7. Appendix 7: Protocol Amendment History

The Protocol Amendment Summary of Changes Table for the current amendment is located directly before the Table of Contents.

Amendment 7 (18 March 2021)

Overall Rationale for the Amendment: To restrict MOPA testing in Cohort 2 to timepoints Day 1, Day 30, Day 181, Year 1, and Year 3 (Day 15 and Year 2 will not be evaluated). It was decided to leave out Day 15 MOPA testing as previous experience with ExPEC4V (63871860BAC2001) showed that Day 15 and Day 30 MOPA results are very similar. MOPA testing at Year 2 was considered not needed as the Applicant believes that, based on experience with ExPEC4V, Year 1 and Year 3 testing are sufficient to model the long-term immunogenicity profile.

Section Number and Name	Description of Change	Brief Rationale
1.1 Synopsis 3.2 Cohort 2 - Double-Blind Period With Double-Blind Long-Term Follow-up Period (N=±420)	MOPA testing at Day 15 in Cohort 2 was deleted.	Previous experience with ExPEC4V (63871860BAC2001) showed that Day 15 MOPA results are very similar to Day 30 MOPA results. The Applicant therefore decided to delete Day 15 MOPA testing.
1.1 Synopsis 3.2 Cohort 2 - Double-Blind Period With Double-Blind Long-Term Follow-up Period (N=±420) 4.1.2 Cohort 2 9.5.2 Cohort 2	MOPA testing at Year 2 in Cohort 2 was deleted.	Based on previous experience with ExPEC4V, MOPA testing at Year 1 and Year 3 are considered sufficient to model the long-term immunogenicity profile.
1.3.2 Cohort 2: Cohort 2: Double-Blind Period Until Day 181 1.3.3 Cohort 1: Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants) 6.5 Concomitant Therapy	Text on concomitant therapy was updated to allow for COVID-19 vaccination.	Because of the global COVID-19 pandemic, a major part of the population will receive a COVID-19 vaccine. The adjustment was made to allow for participants who have received a COVID-19 vaccine or who will receive a COVID-19 vaccine after administration of study vaccination, to be still included in the study.
4.1 Overall Design 10.1 Abbreviations 10.6 COVID-19 Appendix	Incorporation of the stand-alone COVID-19 appendix (14 May 2020) as part of the protocol.	For practical reasons.
Throughout the protocol	Minor formatting errors were corrected.	Minor errors were noted.

Amendment 6 (24 November 2020)

Overall Rationale for the Amendment: To update the number of participants to be included due to early enrollment termination and to clarify that the use of oral *E. coli* vaccines is allowed and must be recorded as concomitant medication.

Section Number and Name	Description of Change	Brief Rationale
1.1 Synopsis 1.2 Schema 3 Objectives and Endpoints	The number of participants to be included in Cohort 2 was updated from 600 to	This update was made based on the decision to prematurely stop participant enrolment.

Section Number and Name	Description of Change	Brief Rationale
3.2 Cohort 2 - Double-Blind Period With Double-Blind Long-Term Follow-up Period (N=±420) 4.1 Overall Design 4.1.2 Cohort 2 8.5 Other Evaluations 9.2 Sample Size Determination	approximately 420. The 2:1 randomization ratio was maintained.	
1.3.2 Cohort 2: Double-Blind Period Until Day 181 1.3.3 Cohort 1 Open-Label Long-Term Follow-up Period (ExPEC10V Selected Dose Group and Prevnar 13 Group) and Cohort 2 Double-Blind Long-Term Follow-up Period (All Participants) 6.5 Concomitant Therapy	A statement was added to clarify that the use of oral licensed *E. coli* vaccines (eg, Uro-Vaxom [OM-89] or Uromune) is allowed for the duration of the study, including long-term follow-up, and must be recorded as concomitant therapy (applicable to European Union countries only).	This clarification was added to be able to determine the clinical/scientific impact, if applicable, of oral licensed *E. coli* vaccines use in the analysis and interpretation of results.

Amendment 5 (15 July 2020)

Overall Rationale for the Amendment: To update the pausing rules criteria to include serious adverse events that are determined to be related to the study vaccine.

Section Number and Name	Description of Change	Brief Rationale
9.7 Study Vaccination Pausing Rules	The pausing rules criteria have been updated to include serious adverse events. In addition, the criterion of 'a persistent Grade 4 laboratory abnormality that is determined to be related to the study vaccine' is now applicable to Cohort 1 and Cohort 2.	Per Health Authority request.

Amendment 4 (17 March 2020)

Overall Rationale for the Amendment: To revise the inclusion criterion for the body mass index range, and to clarify details on data collection for solicited adverse events (AEs) that are not resolved within 14 days after vaccination, severity grading scales, and the time window for participant collection of stool samples.

Section Number and Name	Description of Change	Brief Rationale
5.1. Inclusion Criteria 9.5.2. Cohort 2	The upper limit of the body mass index range for enrollment in the study was revised. Text was added to clarify that the effect of body mass index on immunogenicity will be assessed.	The revised body mass index better reflects the general target population in view of a planned ExPEC10V Phase 3 efficacy study.
8.2. Adverse Events and Serious Adverse Events	Text was updated to clarify that the collection of data on the	The ediary is returned by the participants at the Day 15 visit and is

Section Number and Name	Description of Change	Brief Rationale
	resolution of solicited AEs that are not resolved within 14 days after vaccination will be performed by study-site personnel rather than participants.	not enabled to capture AE recording beyond Day 15.
10.3. Appendix 3. AEs: Definitions and Procedures, Severity Criteria	Text was updated to clarify that all AEs are graded based on the modified FDA toxicity tables in Section 10.5. Redundant severity grading tables for solicited events were deleted.	To clarify severity grading scales applicable to the study. There was no actual change to the severity grading for either Cohort 1 or 2.
1.3.2. SoA, Cohort 2: Double-blind Period Until Day 181 8. Study Assessments and Procedures	The time window during which participants should collect a stool sample before delivery to the study site was clarified.	The time window for participant collection of a stool sample before delivery to the study site was not previously specified in the protocol.
2.1. Study Rationale 2.2. Background	Details regarding two ExPEC4V Phase 2 clinical studies were updated.	The status of ExPEC4V Phase 2 clinical studies has changed since the previous amendment.

Amendment 3 (09 October 2019)

Overall Rationale for the Amendment: To expand the dataset supporting the short- and long-term safety and immunogenicity of the selected dose of ExPEC10V in an additional cohort (Cohort 2). Cohort 2 will include 600 male and female participants ≥60 years of age in stable health with a history of urinary tract infection (UTI) in the past 5 years and will be included in the study to support the plan for late stage development of ExPEC10V.

Section Number and Name	Description of Change	Brief Rationale
1. Protocol Summary 2.1. Study Rationale 3. Objectives and Endpoints 4. Study Design 5. Study Population 6. Study Intervention 7.1.1. Contraindications to Study Vaccination 8. Study Assessments and Procedures 9. Statistical Considerations 10.2. Appendix 2	Study design was updated to add Cohort 2 including participants ≥60 years of age in stable health with a history of UTI in the past 5 years.	Cohort 2 will be included in the study to support the plan for late stage development of ExPEC10V. The Phase 3 target study population will include participants with a history of UTI, which was identified as a key risk factor for developing invasive ExPEC disease based on database mining studies.
1. Protocol Summary 4.1.1. Cohort 1 8. Study Assessments and Procedures 10.2. Appendix 2	Study design was updated to include Prevnar 13 group in the open-label long-term follow-up (LTFU) period in Cohort 1.	Prevnar 13 group will be included as a control group for safety and immunogenicity assessments in the open-label LTFU period in Cohort 1.
1.1. Synopsis 2.1. Study Rationale 3. Objectives and Endpoints 4.1. Overall Design 8. Study Assessments and Procedures 9. Statistical Considerations 10.1. Appendix 1 10.2. Appendix 2	Immunogenicity assays enzyme-linked immunosorbent assay (ELISA) and opsonophagocytic killing assay (OPA) were replaced by multiplex electrochemiluminescent (ECL)-based immunoassay and multiplex opsonophagocytic assay (MOPA), respectively.	For determination of serotype-specific total immunoglobulin G antibodies, the O-antigen- and exotoxin protein A (EPA)-specific singleplex ELISA will be changed to "multiplex ECL-based immunoassay" to support the simultaneous measurement of antibodies against the ten O-antigens and EPA in a high-throughput

Section Number and Name	Description of Change	Brief Rationale
		format. Similarly, the OPA terminology will be changed to MOPA to reflect the change from a singleplex OPA format to a multiplex opsonophagocytic assay that allows measurement of all ExPEC10V serotype-specific functional antibodies.
Throughout the protocol	Wording was changed or added in alignment with revised protocol template.	Due to protocol template changes.
Throughout the protocol	Minor grammatical, formatting, or spelling changes were made, and minor errors and inconsistencies were corrected.	Minor errors were noted.

Amendment 2 (05 April 2019)

Overall Rationale for the Amendment: To update the Phase 1 study design based on Food and Drug Administration (FDA) recommendations and clarify that the additional blood draw from additional 30 participants will be taken from one site in Phase 2a.

Section Number and Name	Description of Change	Brief Rationale
1.1. Synopsis 1.2. Schema 1.3.1. Observer-Blind Period 2.3.5. Overall Benefit/Risk Assessment 4.1 Overall Design 6.3. Measures to Minimize Bias 8. Study Assessments and Procedures 10.2. Appendix 2. Committees Structure (DRC)	Phase 1 study design was updated to reflect a stepwise dose-escalation approach	The study design was updated in line with the FDA recommendation for Phase 1 of the study.
1.3.1. Observer-Blind Period (SoA) 8. Study Assessments and Procedures 8.4. Immunogenicity Assessments	At Day 30, an additional immunogenicity blood draw of 50 mL will be collected from a total of 30 participants (willing to donate blood) in a stepwise manner at each dose level: • Up to 15 participants from low dose and medium dose groups in Phase 1 • Up to 15 participants from high dose group in Phase 1 • If needed, remainder from Phase 2a participants	With updated study design, the text was updated to clarify that additional immunogenicity blood draw of 50 mL will be collected from a total of 30 participants (willing to donate blood) in a stepwise manner at each dose level.
10.3. Appendix 3. Severity Criteria 10.5. Appendix 5. Toxicity Tables	Updated severity criteria and toxicity grading scales for local and systemic adverse events	Due to template changes
Throughout the protocol	Minor abbreviation, grammatical, or formatting changes were made.	Minor errors were noted.

Amendment 1 (21 January 2019)

Overall Rationale for the Amendment: To correct the fever grading (on the impact of fever grading in the eCRF, correct recording of the fever grades).

Section Number and Name	Description of Change	Brief Rationale
1.1. Synopsis 2. Introduction	IED definition update	The text was updated in line with the updated definition of IED and aligned with other documents.
2.2. Background	Study TOX13465 results added	Results from the study TOX13465 were added.
2.2. Background 2.2.1. Comparator Vaccines	63871860BAC2003 study update	The updated status for 63871860BAC2003 was added.
10.5. Appendix 5 Toxicity Tables	Corrected the fever grading/scales in the toxicity tables	The grading scale for fever was corrected to ensure correct recording of fever grades in the eCRF.
Throughout the protocol	Minor abbreviation, grammatical, or formatting changes were made.	Minor errors were noted.

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INVESTIGATOR AGREEMENT

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

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

Coordinating Investigator (where required):

Name (typed or printed): _____

Institution and Address: _____

_____

_____

_____

Signature: _____ Date: _____

(Day Month Year)

Principal (Site) Investigator:

Name (typed or printed): _____

Institution and Address: _____

_____

_____

_____

Telephone Number: _____

Signature: _____ Date: _____

(Day Month Year)

Sponsor's Responsible Medical Officer:

Name (typed or printed): PPD

Institution: Janssen Research & Development, LLC

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

(Day Month Year)

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

Signature

User	Date	Reason
PPD	22-Oct-2021 07:18:59 (GMT)	Document Approval