**VAX24-101**

**Statistical Analysis Plan**: A Phase 1/2a, Randomized, Observer-Blind, Dose-Finding, Controlled, Parallel-Group, Two-Stage Clinical Study to Evaluate the Safety, Tolerability, and Immunogenicity of a 24-Valent Pneumococcal Conjugate Vaccine (VAX-24) in Healthy Adults Aged 18 to 64 Years

**NCT IDENTIFIER:** NCT05266456

**DOCUMENT DATE:** 29 Dec 2022

**Protocol VAX24-101**

**A Phase 1/2a, Randomized, Observer-Blind, Dose-Finding, Controlled, Parallel-Group, Two-Stage Clinical Study to Evaluate the Safety, Tolerability, and Immunogenicity of a 24-Valent Pneumococcal Conjugate Vaccine (VAX-24) in Healthy Adults Aged 18 to 64 Years**

| | |
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| **Protocol Number:**<br>**(Version Date)** | VAX24-101<br>v.4.0 / 31-MAY-2022 |
| **Name of Test Drug:** | VAX-24<br>(24-valent Pneumococcal Conjugate Vaccine) |
| **Phase:** | 1 / 2a |
| **Methodology:** | Randomized, Observer-Blind, Dose-Finding, Controlled, Parallel-Group |
| **Sponsor:** | Vaxcyte, Inc.<br>825 Industrial Road<br>Suite 300<br>San Carlos, CA 94070 USA<br>Tel: 650 837 0111 |
| **Sponsor Representative:** | [redacted]<br>Medical Monitor |
| **Document Date:** | 29-DEC-2022 |
| **Document Version:** | v3.0 |

SIGNATURE PAGE

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|---|---|
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| **Document Date/Version:** | 29-DEC-2022/ Final v3.0 |

[redacted]

**Sponsor Approval**

By signing this document, I acknowledge that I have read the document and approve of the planned pharmacokinetic analyses described herein. I agree that the planned pharmacokinetic analyses are appropriate for this study, are in accordance with the study objectives, and are consistent with the pharmacokinetic methodology described in the protocol, clinical development plan, and all applicable regulatory guidance's and guidelines.

I have discussed any questions I have regarding the contents of this document with the pharmacokinetic author.

I also understand that any subsequent changes to the planned pharmacokinetic analyses, as described herein, may have a regulatory impact and/or result in timeline adjustments. All changes to the planned analyses will be described in the clinical study report (CSR).

[redacted]

[redacted]

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## LIST OF IN-TEXT TABLES

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## ABBREVIATIONS

| | |
|---|---|
| AE | Adverse Event |
| ALT | Alanine Aminotransferase |
| ANCOVA | Analysis of covariance |
| ANOVA | Analysis of variance |
| AST | Aspartate aminotransferase |
| BLQ | Below limit of quantification |
| CI | Confidence Interval |
| CBER | Center for Biologics Evaluation and Research |
| CRF | Case Report Form |
| CSR | Clinical Study Report |
| DBL | Database Lock |
| DMC | Data Monitoring Committee |
| EXP | Exposed Population |
| GMC | Geometric Mean Concentration |
| GMT | Geometric Mean Titer |
| GMFR | Geometric Mean Fold Rise |
| HBsAg | Hepatitis B surface antigen |
| HCV | Hepatitis C Virus |
| HIV | Human Immunodeficiency Virus |
| ICH | International Conference on Harmonisation |
| iDBL | Interim Data Base Lock |
| IEP | Immunogenicity Evaluable Population |
| IgG | Immunoglobulin G |
| IP | Investigational Product |
| LLOQ | Lower Limit Of Quantification |
| MAAE | Medically Assisted Adverse Event |
| MedDRA | Medical Dictionary of Regulatory Activities |
| MCV | Mean Corpuscular Volume |
| NOCI | New Onset of Chronic Illnesses |
| OPA | Opsonophagocytic antibody |
| PCV | Pneumococcal Conjugate Vaccine |
| PD | Protocol Deviation |
| PT | Preferred Term |

| | |
|---|---|
| RND | Randomized Population |
| SAE | Serious Adverse Event |
| SAF | Safety Population |
| SAP | Statistical Analysis Plan |
| SI | International System of Units |
| SOC | System organ class |
| TEAE | Treatment Emergent Adverse Event |
| TLFs | Tables, listings and figures |
| ULOQ | Upper Limit of Quantification |
| VAX-24 | 24-valent investigational pneumococcal conjugate vaccine |
| WHO | World Health Organization |

# 1. INTRODUCTION AND OBJECTIVES OF ANALYSIS

## 1.1. Introduction

This statistical analysis plan (SAP) is designed to outline the methods to be used in the analysis of study data to answer the study objective(s). Analysis sets, data handling rules, statistical methods, and formats for data presentation are provided. The statistical analyses and summary tabulations described in this SAP will provide the basis for the results sections of the clinical study report (CSR) for this trial.

Analyses planned for the Data Monitoring Committee (DMC) meeting at the end of Phase 1 will be described in a separate document.

## 1.2. Objectives

The primary objectives are:

- To evaluate the safety and tolerability of a single injection of VAX-24 at three dose levels administered to healthy adults 18 to 64 years of age.
- To compare the safety of VAX-24 to that of PCV20 administered to two control groups: 1) subjects 18 to 49 years of age receiving PCV20, and 2) subjects 50 to 64 years of age receiving PCV20.

The secondary objectives are:

- Safety: To assess laboratory value abnormalities and/or potentially clinically significant laboratory values following VAX-24 at 3 dose levels compared to control groups receiving PCV20 for subjects aged 50 to 64 years.
- Immunogenicity: To assess the induction of antibody responses by VAX-24 dose levels compared to control groups receiving PCV20 for subjects aged 50 to 64 years.

The exploratory objective is to further characterize the immune response to PCVs in 100 subjects 50 to 64 years of age. Serum samples may also be used for further characterization and validation of clinical assays

# 2. STUDY DESIGN

## 2.1. Introduction

This Phase 1/2a parallel-group, randomized, observer-blind study is to be conducted in two populations of healthy adults, 18 to 49 years of age and 50 to 64 years of age. Subjects will be randomly assigned in a 1:1:1:1 ratio to receive either VAX-24 at one of three dose levels or the active comparator.

A Phase 1 cohort of 64 subjects (16 for each VAX-24 dose group and 16 for the comparator group) aged 18 to 49 years will be enrolled initially. Subjects will receive VAX-24 or PCV20 on Day 1. All subjects will have safety labs analyzed at Baseline and 28 days post-vaccination (Day 29). Solicited adverse events (AEs) will be collected for 7 days post-vaccination and unsolicited safety information for 28 days post-vaccination, with serious adverse events (SAEs), new onset of chronic illness (NOCI) and medically attended adverse events (MAAEs) collected up to 6 months post-vaccination. A defined safety review of data (solicited and unsolicited AEs, and SAEs) through 7 days post-vaccination will occur by an independent DMC before proceeding with enrollment of the remaining subjects, adults 50 to 64 years of age.

Subjects in the 50-to-64-year-old group will receive VAX-24 or PCV20. Solicited and unsolicited AEs will be collected similarly to the subjects aged 18 to 49 years described. All subjects in this age group will have blood samples drawn for immunogenicity analysis (Opsonophagocytic antibody (OPA) and Immunoglobulin G (IgG)) at Days 1, 29.

Approximately 100 of the 800 subjects from 50 to 64 years of age will be consented for collection of additional blood samples (up to 32 mL per draw) by a limited number of clinical sites; subjects will have blood samples drawn at Days 1 and 29 to further characterize the immune response to PCVs.

The schedule of assessments is in Appendix A, further details available in the study protocol.

## 2.2. Sample Size and Power

Sample size is not driven by statistical assumptions.

For subjects 18 to 49 years of age, with 16 subjects, events that occur at a frequency of 15% or more will be detected with at least 92% probability and events that occur at a frequency of 10% or more will be detected with 81% probability. Therefore 16 subjects in each treatment group are proposed as the basis for the number of subjects in whom to conduct the first safety assessment.

For subjects 50 to 64 years of age, a total of 800 subjects will provide sufficient information to assess safety and immunogenicity. The sample size will also be adequate to plan future studies.

## 2.3. **Randomization Methodology**

For the Phase 1 Cohort (18-to-49-year-old age range), a total of 64 subjects are planned to be randomized in a 1:1:1:1 allocation ratio i.e.16 subjects in each VAX-24 dose arm and 16 in the Prevnar group. There will be no stratification factors.

For the Phase 2 Cohort (50-to-64-year-old age range), a total of 800 subjects are planned to be randomized in a 1:1:1:1 allocation ratio i.e.200 subjects in each VAX-24 dose arm and 200 in the Prevnar group. The randomization will be stratified using the following age groups as a stratification factor:

- Age ≥50 and ≤59 years old
- Age ≥60 and ≤64 years old

## 2.4. **Stopping Rules**

The rules governing the halting or stopping of the study vaccine administration at any time during the study are defined by age groups 18 to 49 and 50 to 64 years in the Phase 1 and Phase 2 segments of the clinical study, respectively. During the vaccination period(s), if any of the stopping rules are triggered, no further administration of study vaccine(s) will occur until safety data are reviewed by the DMC in communication with the Sponsor Medical Director and the Investigator at the site where the event occurred, as needed. Study dosing may be resumed if the DMC determines it is safe to proceed with no modifications or with modifications to the protocol plans.

[redacted]

Enrollment may be resumed following review of available safety data by the Sponsor.

The study is planned to be completed after all subjects have completed the Day 180 (Month 6) visit (or Early Discontinuation, as appropriate), all necessary safety follow-up has been completed, and all data have been monitored and queries have been resolved. The Sponsor reserves the right to terminate the study prior to the planned study completion.

## 2.5. **Blinding**

This is an observer-blind study in which all participants, Investigators, and study personnel involved in the conduct of the study, including data management, are blinded to treatment assignment except for:

- Unblinded independent statisticians who will prepare and have access to the randomization code
- Third-Party Vendors (Interactive Response Technology and Supply Management)
- Unblinded pharmacist or designee at site who will prepare Investigational Product (IP)
- Unblinded Administrator at site who will dispense IP
- Unblinded Clinical Research Associate/Study Monitor who will monitor pharmacy
- Unblinded Lead Clinical Research Associate and Clinical Trial Assistant who will review the unblinded Clinical Research Associate/Study Monitor's reports and manage IP supply orders and inventory
- Unblinded Sponsor QA Consultant(s)

Although unblinded to treatment assignment, none of the above roles will have access to unblinded summaries of immunogenicity or safety data.

The following personnel will have access to unblinded data, including individual treatment assignment and individual immunogenicity data, in order to discharge their roles for data analysis and review during the planned Interim Analysis at the end of Phase 1 and the Day 29 Analysis:

- The unblinded statistical programming team, the unblinded data manager, and independent support statistician who will prepare unblinded reports for DMC meetings
- The DMC members who will review unblinded data reports
- The unblinded programming team and the unblinded statistician who will prepare the unblinded report for Day 29 Analysis
- Unblinded Sponsor Reviewer Consultant

Other than the above-mentioned personnel, all other individuals involved in the study conduct, statistical analysis and reporting will remain blinded to individual treatment assignments and individual immunogenicity data until official study unblinding at the end of the study.

Sponsor personnel and limited blinded personnel will become treatment-group unblinded and have access to immunogenicity and safety summaries after the interim database lock (iDBL) for the Day 29 Analysis, as described in Section 2.7.

## 2.6. **Interim Analyses**

A defined safety evaluation will be conducted for the Phase 1 cohort of subjects aged 18 to 49 years, based on the safety data collected through Day 8. The results will be reported to the DMC by treatment group preserving the double-blind status on the subject level, unless the DMC requests unblinded individual treatments. If the interim safety analysis supports and DMC concludes that no safety concerns exist, the study will resume enrollment of subjects aged 50 to 64 years.
For further information including the tables, listings and figures (TLF) to be provided for the DMC review, please refer to the VAX24-101 DMC Charter and associated DMC Mock tables, listings and figures (TLFs) document.

## 2.7. **Schedule of Analyses**

Statistical analyses described in this SAP will be carried out in two stages:

- Day 29 Analysis:
  An interim DBL will occur after all subjects have completed the Day 29 visit and their data up to Day 29 visit have been cleaned. Primary Safety and Immunogenicity endpoints will be produced by programmers with the support of an unblinded statistician. The exact scope and list of outputs for the Day 29 Analysis will be decided prior to the iDBL by the Sponsor and documented in separate TFL shells documents. The results by treatment group will then be available to Sponsor and other study personnel. All blinded personnel, as indicated in Section 2.5, will remain blinded to individual treatment until final database lock. To that effect, no listing of individual data will be part of the Day 29 Delivery and the remaining outputs will be reviewed first by the Unblinded Sponsor Reviewer to assess potential unblinding. The Unblinded Sponsor Reviewer will redact any results by treatment group that may be unblinding and provide the redacted outputs to the wider blinded team.

- Final Analysis:
  The final DBL will occur when all subjects have completed the study, and all data through the last Month 6 visit have been cleaned. Individual Treatment unblinding will be available to all study personnel after the final DBL. The Final Analysis will include all analyses described in this SAP. A single completed Clinical Study Report (CSR) will summarize safety findings through Month 6 for all subjects and immunogenicity data for subjects 50 to 64 years of age.

# 3. STUDY ENDPOINTS

## 3.1. Efficacy Variables

No Efficacy endpoint has been defined for this study.

## 3.2. Safety Variables

Safety assessments performed during the study included solicited AE up to Day 8 and monitoring of unsolicited adverse events, physical examinations, measurement of vital signs, clinical laboratory evaluations including hematology, serum chemistry, and urinalysis.

The primary endpoints are:

- Percentage of subjects reporting solicited local reactions within 7 days after vaccination (redness, swelling, and pain at injection site) in each age group.
- Percentage of subjects reporting solicited systemic events within 7 days after vaccination (fever, headache, fatigue, muscle pain, and joint pain) in each age group.
- Percentage of subjects reporting unsolicited AEs within 1 month after vaccination in each age group.
- Percentage of subjects reporting SAEs, NOCIs, and MAAEs within 6 months after vaccination.

The secondary endpoint related to Safety assessments is:

- Percentage of participants with laboratory value abnormalities and/or potentially clinically significant laboratory values at 1 month after vaccination.

## 3.3. Immunogenicity Variables

Immunogenicity samples will be collected for subjects aged 50 to 64 years only at Day 1, Day 29 and at the Early Discontinuation Visit for subjects discontinuing before Month 6 visit. Immunogenicity parameters to be determined include OPA and IgG assays.

The secondary endpoints related to Immunogenicity assessments are:

- 24 VAX-24 Pneumococcal serotype-specific OPA geometric mean titer (GMTs) at 1 month after vaccination (Day 29).
- 24 VAX-24 Pneumococcal serotype-specific IgG geometric mean concentration (GMCs) at 1 month after vaccination (Day 29).

The exploratory endpoints are all evaluated at 1 month after vaccination (Day 29):

- Geometric Mean Fold Rise (GMFR) in serotype-specific OPA

- GMFR in serotype-specific IgG
- Percentage of participants with GMFR ≥ 4 in serotype-specific OPA
- Percentage of participants with GMFR ≥ 4 in serotype-specific IgG
- Comparison of geometric mean ratio (GMR) of serotype specific OPA and IgG
- Proportion achieving an OPA titer of at least the serotype-specific Lower Limit of Quantitation (LLOQ) of the assay
- Reverse cumulative distribution curves for OPA titers

[redacted]

# 4. ANALYSIS SETS

## 4.1. Analysis Set Definitions

The following Analysis sets will be evaluated and used for presentation and analysis of the data:

- Screened Population: Includes all screened subjects who provided informed consent and were assigned a study subject number, regardless of whether the subject was randomized or not. This population will be used to account fully for subject disposition, starting with the informed consent. The screened population will not be analyzed as such but will be available in the clinical database.
- Randomized Population (RND): Includes all subjects from the Screened Population who consented, provided demographic and other Baseline Screening measurements, and were randomized. Each subject will be analyzed as randomized.
- Exposed Population (EXP): Includes all subjects from the Screened Population who received at least one study vaccine administration. Each subject will be analyzed as treated.
- Safety Population (SAF): Includes all subjects in the Exposed Population but excluding subjects lost to follow up at Day 1 reporting no solicited or unsolicited AEs. Each subject will be analyzed as treated.
- Immunogenicity Evaluable Population (IEP): Includes all subjects in the Exposed Population who:
    - Had no major protocol deviation that would impact immunogenicity assessment or other reason to be excluded as defined prior to unblinding or analysis.
    - Had not received a prohibited medication or vaccine. Identification of subjects receiving prohibited medications will be done via a medical review from the Sponsor of a listing of all concomitant medications.
    - Provided evaluable serum sample results for baseline, the relevant post-vaccination time points, and within the required time frames:
        - Baseline: Day 1 or within 30 days before first study vaccine administration
        - Day 29: Day 26 through Day 34, inclusive

  Each subject will be analyzed as treated.

"As randomized" means according to the vaccine regimen to which the subject was randomized, while "as treated" means according to the vaccine regimen a subject received, rather than the vaccine regimen to which the subject may have been randomized.

The RND will be used for the analysis of exposure.
The SAF will be the primary set for the analysis of the safety parameters.
The IEP will be the primary set for the immunogenicity analysis, with the EXP set as supportive.

## 4.2. **Protocol Deviations**

All deviations from the protocol are documented in the study file. In addition, deviations are reported to the Institutional Review Board as applicable.

Subject-specific deviations are recorded in the subject's source documents. The Sponsor will review all protocol deviations (PDs) on an ongoing basis and will be responsible for categorizing protocol deviations as exclusionary from IEP. Before the iDBL and the Final DBL, [redacted] will download the 'dv' csv file from Encapsia including all deviations entered in the system and will send it to the Sponsor. The Sponsor will use this source data to produce a classification Excel file by adding 2 new columns:

- IEPEXCL ("Leads to Exclusion from IEP?"). This column will be filled for each PD with:
    - 'Yes' if the PD leads to exclusion of the subject from the IEP
    - 'No' if the PD does not lead to exclusion of the subject from the IEP
- IEPEXCLREAS ("Reason for leading to exclusion from IEP"). This column will be filled for each row where IEPEXCL=Yes, with some categorical reasons of exclusions. Since prohibited medication/vaccine, immunogenicity samples taken out of window, and other major PDs will be recorded as PD and classified as exclusionary from IEP by the Sponsor, only the variable IEPEXCL will be used by [redacted] to identify subjects excluded from IEP.

The PD classification file will be finalized prior to iDBL for the Day 29 Analysis, and prior to the final DBL for the Final Analysis.

All PDs and their classification will be presented in a data listing.

# 5. DATA HANDLING

## 5.1. Computing Environment

All descriptive statistical analyses will be performed using SAS statistical software (Version 9.4), unless otherwise noted. Medical History and adverse events will be coded using Medical Dictionary of Regulatory Activities (MedDRA) version 24.1. Concomitant medications will be coded using B3 World Health Organization (WHO) Drug Global (September 2021).

## 5.2. Data Conventions

The following conventions will be used:

- **Period definition:**
  - **Screening Period:** The period prior to Day 1 visit.
  - **Treatment and Observation:** The period from date of vaccine administration to Day 29 visit.
  - **Follow-up:** The period from Day 29 to Month 6 visits.
- **Visits:**
  - **Study day 1:** The date of study vaccine administration.
  - **End of Treatment Visit:** The Day 29 visit, or the early discontinuation visit for subjects who withdraw study prior to Day 29.
  - **End of Study Visit:** The last recorded visit date.
  - **Unscheduled visits:** Unscheduled visits results will be listed, but not included in tables or graphs. Rules to map unscheduled visit to analysis visit are defined in Section 5.5.
- **Conversion factors (for derived data calculations):**
  - 1 month = 30.4375 days
  - 1 week = 7 days
- **Baseline characteristics and change from baseline:**
  - Weight values recorded in pounds will be converted to kilograms using the following formula: kilograms = pounds/2.2046.
  - Height values recorded in inches will be converted to centimeters using the following formula: centimeters = inches*2.54.
  - Duration on study (weeks) = (Last visit date – randomization date + 1) / 7.
  - (Absolute) Change from baseline = Value at the time point – Baseline value.

- **Adverse events:**
  - **Solicited Adverse Events** will be analyzed using the nominal visit (i.e. the page) on which they were recorded and not according to the date they were entered.
  - **Solicited Adverse Events continuing beyond 7 days**: Solicited AEs continuing beyond 7 days will be reported in the "Adverse Evens (Unsolicited)" page as per Case Report Form (CRF) Completion guidelines and will be identified programmatically as all unsolicited adverse events with answer 'Yes' to the question 'Was this adverse event continuing as reported on the Day 7 subject diary?'.
- **Immunogenicity:**
  - **GMC [or GMT]**: The geometric mean concentration (or titer) = antilog (LSMeans [log10 x]), where x is the assay result and LSMeans is the least square means calculated by the model.
  - **GMR**: The geometric mean ratio = antilog (LSMeans [log10 x]) / antilog(LSMeans [log10 y]), where x and y are the 2 assays results and LSMeans is the least square means calculated by the model.
  - **Fold Ratio**: The ratio between titer/concentration at Day 29 visit and the one at Day 1.
  - **GMFR**: The geometric mean fold ratio = antilog (LSMeans [log10 y]), where y is the assay fold ratio and LSMeans is the least square means calculated by the model.
  - **4-fold increase**: A subject achieved 4-fold increase if fold ratio ≥ 4
  - **BLQ**: Below lower limit of quantification (LLOQ). Per Center for Biologics Evaluation and Research (CBER) criterion, BLQ titer/concentration will be analyzed as:
    - 0.5*LLOQ for calculations of GMT/GMC and GMT/GMC Ratios (GMRs)
    - 1*LLOQ for calculations of fold ratio.
  - **ULOQ**: Upper limit of quantification. Titer/Concentration above ULOQ will be analyzed as:
    - 1*ULOQ for all calculations (GMT/GMC, GMRs, fold-ratio)

## 5.3. **Methods of Pooling Data**

Not Applicable.

## 5.4. **Withdrawals, Dropouts, Loss to Follow-up**

Subjects who undergo Early Discontinuation after randomization and before Day 1 vaccine administration may be replaced by randomizing an additional subject at the Sponsor's discretion. Subjects who undergo Early Discontinuation after study vaccination will not be replaced.

[redacted]

# 6. STATISTICAL METHODS

## 6.1. Sample Size Justification

Sample size is not driven by statistical assumptions for formal hypothesis testing.

For subjects 18 to 49 years of age, with 16 subjects, events that occur at a frequency of 15% or more will be detected with at least 92% probability and events that occur at a frequency of 10% or more will be detected with 81% probability. Therefore 16 subjects in each treatment group are proposed as the basis for the number of subjects in whom to conduct the first safety assessment.

## 6.2. General Statistical Methods

### 6.2.1. General Methods

All outputs will be incorporated into Microsoft Word files, sorted, and labeled according to the International Conference on Harmonization (ICH) recommendations, and formatted to the appropriate page size(s).

Tabulations will be produced for appropriate demographic, baseline, safety, and immunogenicity parameters. For categorical variables, summary tabulations of the number and percentage within each category (with the number of subjects with missing data) of the parameter will be presented. For continuous variables, the mean, median, standard deviation, minimum and maximum values will be presented.

Formal statistical hypothesis testing will be performed on immunogenicity parameters with all tests conducted at the 2-sided, 0.05 level of significance. Summary statistics will be presented, as well as 95% confidence intervals (CI) on selected parameters, as described in the sections below.

### 6.2.2. Definition of Baseline

For all endpoints, baseline is defined as the last non-missing assessment prior to study vaccine administration. Specifically, as per the protocol, baseline evaluations are expected as follows:

- Vital signs: Day 1 prior to study vaccine administration
- Laboratory Parameters: Most recent value during Screening period (Day -30 to Day -1)
- Immunogenicity: Day 1 prior to study vaccine administration

[redacted]

### 6.2.3. Adjustments for Covariates

The randomization for Phase 1 Cohort will not be stratified.

The randomization for Phase 2 Cohort will be stratified by age group (50-59, 60-64).

Immunogenicity analyses:

- For the Analysis of variance (ANOVA), study site will be included as fixed effect in the model.
- For the Analysis of covariance (ANCOVA) and the logistic regression, study site will be included as fixed effect and the log baseline titer will be included as covariate in the model.

### 6.2.4. **Multiple Comparisons/Multiplicity**

Since there are no formal hypothesis tests in this study, no adjustment for multiplicity will be made.

[redacted]

### 6.2.6. **Missing, Unused, and Spurious Data**

Binary endpoints / Continuous data:

Missing data are assumed to be missing at random and ignorable. Missing data will not be estimated or imputed. Denominators for percentages will be based only on the number of subjects with non-missing values. The only exception is:

- Systemic Solicited AEs with missing “Relationship to study vaccine” will be analyzed as “Possibly related”.

Dates:

For partial or missing AE start dates the following imputation rules will be applied:

1. If year is not missing and is after the year of first dose:
    a. If month is missing, then month will be imputed as January.
    b. If day is missing, then day will be imputed as the first of the month.

2. If year is not missing and is the same as the year of the first dose:
    a. If month is missing, then impute the month as the month of the first dose date.
    b. If day is missing, and the month is the same as the month of the first dose date, then impute day as the day of the first dose date.
    c. If day is missing but month is after the month of first dose date, then impute day as the first day of the month.
3. If year is missing, then impute the year as the year of the first dose date:
    a. If month is missing, then impute the month as the month of the first dose date.
    b. If day is missing, then impute the day as the day of the first dose date.
4. If the start date is completely missing, but the AE is either ongoing (i.e. AE stop date is missing) or the stop date is after the first dose date then impute the start date as the first dose date.
5. For any cases involving the rules above, if the AE end date is before the AE start date, then do not impute the AE start date and assume that the AE is treatment emergent/concomitant for the purpose of the analysis. Further, if the AE stop date occurs prior to the first dose date, do not impute the AE start date, and assume that the AE is not treatment emergent.

No imputations will be applied to AE stop dates or other dates. As indicated above, AEs with missing stop date are considered ongoing.

## 6.3. **Study Population**

### 6.3.1. **Subject Disposition**

Subject disposition will be presented by treatment group and overall, including the number screened, the number of screen failures, the number randomized and the number who received study vaccine administration. The study disposition will also be presented along with the reasons for early study withdrawal.

The number of subjects in each analysis set will be presented on the Randomized Population by treatment group and overall.

The following by-subject listings will be presented.

- Study completion information, including the reason for premature study withdrawal
- Visit dates
- Inclusion in study Analysis Sets and reason for exclusion from the IEP
- Protocol deviations

### 6.3.2. **Demographic Characteristics**

Demographic characteristics at enrollment (i.e., at the time of informed consent) will be presented. Summary will include the following: age, sex, ethnicity, race, height, weight, body mass index.

Medical history will be coded using Medical Dictionary for Regulatory Activities (MedDRA) and summarized alphabetically by System Organ Class (SOC) and Preferred Term (PT).

Childbearing potential and method of contraception will be presented in Listings only.

Table summaries to be produced by treatment group and overall and repeated for the following:

Populations: RND, SAF, IEP

Subgroups: 18-64, 18-49, 50-64

Demographic characteristics and medical history data will also be provided in data listings on the Randomized Population.

### 6.3.3. **Prior and Concomitant Medication**

Prior and concomitant medications will be coded using the WHO Drug dictionary.

Prior medications will be defined as any medications with a start date before the date of study vaccine administration.

Concomitant medications will be defined as any medications with a start date on or after the date of study vaccine administration, as well as medications taken prior to the study vaccine administration and continuing after.

If a medication date or time is missing, or partially missing, and it cannot be determined whether it was taken on or after start of treatment, it will be considered a concomitant medication.

Results will be tabulated by Anatomic Therapeutic Class level 2 and preferred term.

Table summaries to be produced by treatment group and overall and repeated for the following:

Population: SAF

Subgroups: 18-64, 18-49, 50-64

Prior and concomitant medications will be included in a by-subject data listings.

### 6.3.4. **Exposure and Compliance**

Frequency and percentage of subjects with vaccinations will be summarized. For subjects who received a vaccine, the administered vaccine will be summarized.

Table summary to be produced by treatment group and repeated for the following:

Population: RND

Subgroups: 18-64, 18-49, 50-64

## 6.4. **Efficacy Evaluation**

Not Applicable.

## 6.5. **Safety Evaluations**

All Safety summaries to be produced by treatment group, for the following:

Population: SAF

Subgroups: 18-64, 18-49, 50-64

Summaries including primary endpoints (noted as [P]), secondary endpoints (noted as [S]) or exploratory endpoints (noted as [E]), will also be repeated for the following:

Population: SAF

Subgroups: 50-59, 60-64

### 6.5.1. **Adverse Events**

#### **6.5.1.1. Solicited Adverse Events**

Solicited AEs are protocol-specified local and systemic symptoms/events that are proactively collected from the subject and evaluated by the Investigator or designee. Solicited AEs are collected for 7 days after each injection, starting on Day 1, within an electronic diary. All solicited AEs will be considered "Probably related" to study vaccine.

Solicited AEs for this study are:

- Local events: Pain, Erythema (redness) at the injection site, Edema (swelling) at the injection site
- Systemic events: Fever (oral temperature ≥ 100.4ºF), Fatigue, Headache, Muscle pain, Joint pain

The Subject and the Investigator will grade all AEs for severity from "mild" (grade 1) to "potentially life-threatening" (grade 4), except for severity of redness and swelling recorded as diameters (cm) and graded accordingly (see Guidance for Industry Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials, September 2007 for details on grading scale).

Only Solicited AEs collected in the eDiary CRF, within 7 days after vaccine administration (i.e. from Day 1 to Day 7) will be analyzed in this Section. Occurrence of a solicited AE on a specific Day will be identified by a Grade≥1 for that AE on that day.

Solicited AEs continuing beyond 7 days after injection will be combined with unsolicited AEs and described separately as specified in Section 6.5.1.3.

The following characteristics will be calculated:

- Maximum severity (Investigator):

For each solicited AE collected for a subject, the maximum severity will be the highest grade recorded, as per Investigator, for all occurrences of this AE within 7 days of vaccine administration (i.e., from Day 1 post-administration to Day 7) for the overall summary, or within a single day for the summary by day. There will be no replacement in case of missing Grade.

- Maximum relationship (Investigator):

  For each unsolicited AE collected for a subject, the maximum relationship will be the strongest relationship, as per Investigator, for all occurrences of this AE. There will be no replacement in case of missing relationship.

- Time of first onset of first event (days):

  Day of onset of the first solicited AE, as per the nominal visit where the first solicited AE was first reported. This will range from 1 (Day 1 = Day of study vaccine administration) to 7 (Day 7).

- Duration of solicited AEs (days):

  Number of days between the onset of the solicited AE (i.e. nominal visit of first occurrence of Grade≥1) and the end of the solicited AE (i.e. nominal visit when Grade is back to 0).
  If there are different occurrences of a same solicited AE within 7 days, the durations will be added up.
  For solicited AEs continuing beyond 7 days, the end day of the continuing event will be used as end day for the calculation.
  *Example: If a subject has a Fatigue starting on Day 2 and finishing on Day 3. And a new Fatigue starting on Day 7 and ending on Day 9. The Duration will be 1+2=3.*
  If the solicited AE is ongoing (i.e. stop date is missing), then duration will be the Number of days between the onset of the solicited AE and the study discontinuation date.

Frequencies and percentages of subjects experiencing each solicited AE will be presented overall and by maximum severity. Summary tables showing the occurrence of any local or systemic solicited AE overall and at each time point will also be presented. For each of the time points or time intervals presented in the summaries, only subjects with at least one observation (ie, any non-missing values but excluding “Not done/unknown”) for the solicited AEs will be summarized.

The following summaries of subjects will be performed:

- [P] Solicited AEs within 7 days post-injection, for each event and for any event, overall and by maximum severity. Details by category (local, systemic) will also be presented. (Severity as per Investigator)
- Solicited AEs by day post-injection, for each event and for any event, overall and by maximum severity on that day. (Severity as per Investigator)
- Solicited AEs by day post-injection, for each event and for any event, overall and by maximum severity on that day. (Severity as per Subject)

- Time of first onset of solicited AEs within 7 days post-injection for each event and any event. Duration of solicited AEs, after injection, for each event. Maximum duration of any event. (Severity as per Investigator)

Listings of all solicited AEs will be provided by subject.

#### 6.5.1.2. Unsolicited Adverse Events

An unsolicited AE is an AE that is spontaneously reported by the subject or discovered by the Investigator. New onset of chronic illness(s) would be recorded as part of the collection of unsolicited AEs as specified in the visit procedures. Unsolicited AEs are collected separately from solicited AEs.

Solicited AEs continuing beyond 7 days will be flagged using rules given in Section 5.2 and described in separate summaries as explained in Section 6.5.1.3.

The definition of a treatment emergent AE (TEAE) is an event that occurs after vaccination and within the 28 days after vaccination (i.e., excluding those after a subject has given informed consent, but before vaccination):

- Onset (days) = Start date of AE – Date of Study Vaccine Administration + 1
- An AE is a TEAE if:
    - Onset≥1 and Onset≤28
    - For AEs with Onset=1 the time of onset of AE must be greater or equal to the time of vaccination to be considered a TEAE

Adverse events are summarized by subject, therefore, in any tabulation, a subject contributes only once to the count for a given adverse event (SOC or preferred term).

Unsolicited AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) and will be summarized by decreasing frequency of SOC and preferred term within SOC in the control group, as follows:

- [P] Any unsolicited TEAEs
- Possibly or probably related unsolicited TEAEs
- Unsolicited TEAEs leading to study withdrawal
- Any AE leading to death
- [P] Any SAE within 6 months after the vaccination by maximum relationship to study treatment. This summary will include serious solicited AEs.
- [P] Any NOCI within 6 months after the vaccination by maximum relationship to study treatment.
- [P] Any MAAEs within 6 months after the vaccination by maximum relationship to study treatment.

Following by-subject Listings will be provided:

- Pre-vaccination AEs (non-emergent)

- All unsolicited TEAEs
- All Serious post-vaccination AEs, NOCIs and MAAEs

#### 6.5.1.3. Combined Solicited and Unsolicited Adverse Events

Solicited AEs continuing beyond 7 days will be recorded by the sites and flagged using rules given in Section 5.2. They will be coded by MedDRA and following summary by decreasing frequency of SOC and preferred term within SOC will be provided:

- Solicited TEAEs continuing beyond 7 days
- Combined solicited TEAEs continuing beyond 7 days and unsolicited TEAEs

Following by-subject Listings will be provided:

- All solicited TEAEs continuing beyond 7 days

### 6.5.2. Laboratory Data

- A summary describing pregnancy tests results, as well as results for Human Immunodeficiency Virus (HIV), Hepatitis B surface antigen (HBsAg) and Hepatitis C Virus (HCV) tests at Baseline will be presented. The list of Baseline test results to be described is given in *Table 2*. Reflex testing (HBsAg confirmation, HCV RNA qual, HIV1AB, HIV2AB) will be described only for subjects with initial positive test.

***Table 2 Baseline Laboratory Tests***

| Panel | Sample | Parameter | Screening | Day 1 |
|---|---|---|---|---|
| Immunochemistry | Serum | Pregnancy (serum) | X | |
| Immunochemistry | Serum | HBsAg | X | |
| Immunochemistry | Serum | HBsAg confirmation | X | |
| Immunochemistry | Serum | HCV Ab | X | |
| Immunochemistry | Serum | HCV RNA qual | X | |
| Immunochemistry | Serum | HIV 1/2 Ab-p24 Ag | X | |
| Immunochemistry | Serum | HIV1AB | X | |
| Immunochemistry | Serum | HIV2AB | X | |
| Urinalysis | Urine | Pregnancy (urine) | | X |

- The actual value and change from Baseline to Day 29 and to Early Study Discontinuation will be summarized for each clinical laboratory parameter, including hematology, clinical chemistry, and urinalysis. The complete lists of parameters can be found in *Table 3*.
- [S] A shift table from Baseline to Day 29 and to Early Study Discontinuation presenting abnormal values for each parameter will also be presented.
  Clinical laboratory values will be expressed using conventional international system of units (SI). In the event of repeat values, the last non-missing value per study day/time will be used. Clinically significant out-of-range lab results will be reported as adverse events by Investigators. Clinical significance will therefore not be reported in the laboratory outputs.

***Table 3 Laboratory Parameters Collected during the Study***

| Panel | Sample | Parameter | Screening | Day 29 (or Early Study Discontinuation) |
|---|---|---|---|---|
| Hematology | Blood | Hemoglobin | X | X |
| Hematology | Blood | White blood cell count (absolute) | X | X |
| Hematology | Blood | Neutrophils (absolute) | X | X |
| Hematology | Blood | Eosinophils (absolute) | X | X |
| Hematology | Blood | Platelet count | X | X |
| Chemistry | Serum | Albumin | X | X |
| Chemistry | Serum | Alkaline phosphatase | X | X |
| Chemistry | Serum | Alanine aminotransferase (ALT) | X | X |
| Chemistry | Serum | Aspartate aminotransferase (AST) | X | X |
| Chemistry | Serum | Bilirubin (Total) | X | X |
| Chemistry | Serum | Bilirubin (Direct) | X | X |
| Chemistry | Serum | Blood urea nitrogen | X | X |
| Chemistry | Serum | Calcium | X | X |
| Chemistry | Serum | Carbon dioxide | X | X |
| Chemistry | Serum | Chloride | X | X |
| Chemistry | Serum | Cholesterol and triglycerides | X | X |
| Chemistry | Serum | Creatinine | X | X |
| Chemistry | Serum | Glucose (random) | X | X |
| Chemistry | Serum | Gamma-glutamyl transferase (GGT) | X | X |
| Chemistry | Serum | Lactate dehydrogenase | X | X |
| Chemistry | Serum | Phosphate | X | X |
| Chemistry | Serum | Potassium | X | X |
| Chemistry | Serum | Sodium | X | X |
| Chemistry | Serum | Total serum protein | X | X |
| Chemistry | Serum | Uric acid | X | X |
| Urinalysis | Urine | Specific Gravity (SG) | X | X |
| Urinalysis | Urine | pH | X | X |
| Urinalysis | Urine | Glucose | X | X |
| Urinalysis | Urine | Protein (Total) | X | X |
| Urinalysis | Urine | Ketones | X | X |
| Urinalysis | Urine | Bilirubin | X | X |
| Urinalysis | Urine | Urobilinogen | X | X |
| Urinalysis | Urine | Hemoglobin | X | X |
| Urinalysis | Urine | Leucocyte Esterase | X | X |
| Urinalysis | Urine | Nitrite | X | X |

All laboratory data will be provided in data listings.

### 6.5.3. Vital Signs and Physical Examinations

The actual value and change from before study vaccine administration to after study vaccine administration will be summarized for each vital sign parameter: Systolic Blood Pressure, Diastolic Blood Pressure, Heart Rate, Oral Temperature, Respiratory Rate. In case of multiple measurements on Day 1 before vaccination, the closest measure from the time of vaccination will be used. In case of multiple measurements on Day 1 after vaccination, the closest measure from the time of vaccination will be used. Vital sign measurements will be presented in by-subject data listings.

Physical examination results at Baseline will be summarized. All physical examination findings will be presented in by-subject data listing.

## 6.6. **Immunogenicity Evaluations (50 to 64 years)**

All Immunogenicity summaries to be produced by treatment group, for the following:

Population: IEP, EXP

Subgroups: 50-64, 50-59, 60-64

### 6.6.1. **GMT/GMC, GMR and GMFR**

To estimate the immunogenicity response 1 month first vaccination, OPA titers (log10) and IgG titers (mcg/mL) measured at Day 29 visit will be logarithmically transformed for analysis and GMTs/GMCs will be computed with 95% CI for each assay using ANOVA and ANCOVA.

- [S] [E] First model in an ANOVA with log10-transformed concentrations/titers at Day 29 as the dependent variable and treatment group and study site as the fixed effects in the model. Bar plots of OPA GMT, IgG GMC and their 95% CI computed by this model will be created for the full IEP only.
- [S] [E] Second model in an ANCOVA with log10-transformed concentrations/titers at Day 29 as the dependent variable, treatment group and study site as the fixed effects and log10 baseline concentration/titer as the covariate

The least squares means, and their 95% CIs calculated based on the ANOVA and ANCOVA will be back transformed and reported as the group GMT and GMC values. Comparisons between relevant groups will be based on the estimated adjusted GMTs measured at Day 29 visit for 24 serotypes in VAX-24 (of those 20 in PCV20), and mean square error calculated from the basic ANOVA model using contrast statements. The main comparison of interest will be the three VAX-24 dose levels group versus the PCV20 (20 serotypes) group. However, the three VAX-24 dose groups will also be compared in a pairwise fashion on a serotype-by-serotype basis. No adjustment for multiplicity will be applied and missing data will not be imputed. GMRs will also be calculated from the ANCOVA and ANOVA models.

The analysis of GMFR at Day 29 relative to Day 1 will also be computed using similar models:

- [E] First model in an ANOVA with log10-transformed fold ratio at Day 29 as dependent variable and treatment group and study site as the fixed effects in the model. A Forest Plot summarizing the results by serotype of this model will be created for OPA on the full IEP only.
- [E] Second model in an ANCOVA with log10-transformed fold ratio at Day 29 as dependent variable, treatment group and study site as the fixed effects and log10 baseline concentration/titer as the covariate

The least squares means, and their 95% CIs calculated based on the ANOVA and ANCOVA will be back transformed and reported as the group GMFR value.

[redacted]

[redacted]

# 7. CHANGES TO PLANNED ANALYSES

As of this date, there have been no changes between the protocol-defined statistical analyses and those presented in this statistical plan.

# 8. REFERENCES

1. International Council on Harmonization, Statistical Principles for Clinical Trials (ICH E9)
2. Guidance for Industry Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials, September 2007

# 9. APPENDICES

## 9.1. Schedule of Assessments

**APPENDIX A: SCHEDULE OF EVENTS**

| | Visit Number: | -- | 01 | 02 | 03 | 04 | 05 - 08 | -- |
|---|---|---|---|---|---|---|---|---|
| | Study Visit: | Screening | Day 1 | Day 8 | Day 15 (Phone) | Day 29 | Months 3, 4, 5, 6 (Phone) | Early Termination |
| Study Event | Window: | -30d | | +3d | +3d | ±3d | ±5d | |
| Informed Consent | | X | | | | | | |
| Demographics, Medical History | | X | X[a] | | | | | |
| Concomitant Medications | | X | X | X | X | X[c] | X[c] | |
| Physical Exam(s), targeted | | X | X[a] | | | | | |
| Vital Signs | | X | X[b] | | | | | |
| Confirmation of Eligibility | | X | X | | | | | |
| Randomization | | | X | | | | | Refer to Protocol for Procedures |
| Study Vaccine Administration | | | X | | | | | |
| Post-vaccination Observation (at least 30 min) | | | X | | | | | |
| Issue e-Diary instructions, Ruler, Thermometer; Conduct Training | | | X | | | | | |
| Review e-Diary Data | | | | X | | | | |
| AE Evaluation (Solicited and/or Unsolicited) | | | X | X | X | X | X[c] | |
| Clinical Labs[d] | | | | | | | | |
| Hematology, Chemistry, Urinalysis Tests | | X | | | | X | | |
| Pregnancy Tests (Urine or Serum)[e] | | X | X | | | | | |
| HIV EIA, HBsAg, HCV Ab | | X | | | | | | |
| Serum for Immunogenicity, [redacted] (50 to 64 yo only) | | | X | | | X | | |

Abbreviations: AE = adverse event; HBsAg = Hepatitis B surface antigen; HCV = Hepatitis C virus antibody; HIV EIA = human immunodeficiency virus, enzyme immunoassay; [redacted] SAE = serious adverse event; yo = years old

[a] Conduct or collect prior to study vaccination (if indicated by updated medical history or change in health status, as applicable)

[b] Vitals to be taken prior and after study vaccine administration (≥ 30 min)

[c] New onset of chronic illnesses, MAAEs and SAEs, and associated concomitant medications collected after Day 29

[d] All samples collected prior to study vaccine administration

[e] Serum pregnancy test at Screening and urine pregnancy test prior to vaccine administration