Janssen Vaccines & Prevention B.V.*

Clinical Protocol

Protocol Title

A Randomized, Double-blind, Phase 2 Study to Evaluate the Immunogenicity, Reactogenicity and Safety of Ad26.COV2.S Administered as Booster Vaccination in Adults 18 Years of Age and Older Who Have Previously Received Primary Vaccination with Ad26.COV2.S or BNT162b2.

Amplify

Protocol VAC31518COV2008; Phase 2

AMENDMENT 6

VAC31518 (JNJ-78436735 [Ad26.COV2.S])

*Janssen Vaccines & Prevention B.V. is a Janssen pharmaceutical company of Johnson & Johnson and is hereafter referred to as the sponsor of the study. The sponsor is identified on the Contact Information page that accompanies the protocol.

United States (US) sites of this study will be conducted under US Food & Drug Administration Investigational New Drug (IND) regulations (21 CFR Part 312).

Regulatory Agency Identifier Number(s):

IND: 22657
EudraCT NUMBER: not applicable

Status: Approved
Date: 20 April 2022
EDMS number: EDMS-RIM-508722, 7.0

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 6	**This document**
Amendment 5	**17 December 2021**
Amendment 4	30 November 2021
Amendment 3	08 October 2021
Amendment 2	22 September 2021
Amendment 1	16 August 2021
Original Protocol	30 June 2021

Amendment 6 (This document)

Overall rationale for the Amendment: This amendment is written primarily to clarify the process for additional interim analyses and final analysis, potential exploratory efficacy analysis, clinical adjudication of COVID-19 disease severity and management of COVID-19 episodes. In addition, this amendment will allow unblinding for participants rolling over into study VAC31518COV2015.

Section Number and Name	Description of Change	Brief Rationale
1.1 Synopsis 9.5 Planned Analyses	Text has been added to allow additional interim analyses to be conducted after the primary analysis.	To allow additional analysis to be performed between the primary analysis and final analysis, allowing earlier evaluation of booster response durability.
1.3.1 Schedule of Activities: All Participants 1.3.3 Schedule of Activities for Participants with COVID-19-like Signs and Symptoms.	The frequency of the COVID-19 surveillance (symptom check), after the 6-month study visit has been changed to at least twice a month.	To reduce burden on trial participants and improve participant retention.
1.1 Synopsis 2.3.3 Benefit-Risk Assessment of Study Participation 4.1 Overall Design 6.8 Study Pausing Rules 9.5 Planned Analysis 10.3.6 Committee Structure	IDMC review of safety has been changed from continual to an ad hoc basis.	As the safety of Ad26.COV2.S is established, it is not necessary for all data to be reviewed by the IDMC from this point onwards (post primary analysis). Emergent safety concerns can be brought to the IDMC at any time.
1.1 Synopsis 3. Objectives and Endpoints 8.2 Exploratory Efficacy Analysis 9.3 Populations for Analysis Sets 9.4.4 Exploratory Endpoints 9.5 Planned Analyses	Text has been added on exploratory efficacy analyses, if sufficient data is available.	To allow exploratory analyses of booster vaccine efficacy after the primary analysis (utilizing molecularly confirmed SARS-CoV-2 infections, N protein binding antibodies, COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition and cases meeting criteria for moderate and moderate/severe disease).

1.1 Synopsis 3. Objectives and Endpoints 9.4.5 Other Analysis	Text has been added on correlate analysis.	To determine immunological correlates of risk and protection, if sufficient data are available.
1.1 Synopsis 3. Objectives and Endpoints 8.1.4 Clinical Severity Adjudication Committee 8.2 Exploratory Efficacy Analysis 10.3.6 Committee Structure	Text on the 'Clinical Severity Adjudication Committee' has been added.	To describe the process of COVID-19 disease severity evaluation in study participants.
1.3.3 Schedule of Activities for Participants with COVID-19-like Signs and Symptoms. 8.1.2 Procedures in the Event of COVID 19-like Signs and Symptoms 8.3 Safety Assessments 8.4.1 Time Period and Frequency for Collecting AEs	Text has been added to clarify that the end of study visit should not be postponed for participants with an ongoing COVID-19 episode.	The End of Trial (EoT) visit should not be postponed in the event a participant has an ongoing COVID-19 illness. The COVID-19 Signs and Symptoms Schedule of Activities can be stopped at the time of the EoT visit.
8.5 Virology Assessments 10.2 Appendix 2: Clinical Laboratory Tests	Wording on saliva samples has been changed from 'will also be performed' to 'may also be performed'.	Testing of viral load in saliva samples will be conducted if feasible.
1.1 Synopsis 6.3 Randomization and Blinding	Edits have been made to the sections relating blinding/unblinding and subsequent enrollment into study VAC31518COV2015.	To permit rollover from study VAC31518COV2008 into Janssen-sponsored study VAC31518COV2015.
5.2 Exclusion Criteria	Text discouraging participation in other trials has been deleted from criterion #7.	The sponsor would like to allow rollover of eligible participants from COV2008 into other Janssen sponsored COVID-19 vaccine studies i.e. VAC31518COV2015.
6.8 Pausing Rules	Text on study pause of COV2007 has been deleted.	This text is obsolete as the protocol for COV2007 was not finalized and the study will not be initiated.
Throughout the protocol	Minor formatting and grammatical changes were made.	Minor errors were noted.

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1. PROTOCOL SUMMARY

1.1. Synopsis

A Randomized, Double-blind, Phase 2 Study to Evaluate the Immunogenicity, Reactogenicity and Safety of Ad26.COV2.S Administered as Booster Vaccination in Adults 18 Years of Age and Older Who Have Previously Received Primary Vaccination with Ad26.COV2.S or BNT162b2.

Ad26.COV2.S (also known as VAC31518, JNJ-78436735) is a monovalent vaccine composed of a recombinant, replication-incompetent adenovirus type 26 (Ad26) vector, constructed to encode the Spike (S) protein derived from a SARS-CoV-2 clinical isolate (Wuhan 2019, whole genome sequence NC 045512, further referred to as the original strain), stabilized in its prefusion conformation.

The Pfizer-BioNTech COVID-19 vaccine, BNT162b2, is a lipid nanoparticle-formulated, nucleoside-modified RNA vaccine encoding a prefusion stabilized, membrane-anchored SARS-CoV-2 full-length spike protein.

In Cohort 1, participants who previously received Ad26.COV2.S primary vaccination in Janssen-sponsored study COV3001, will receive a 1-dose booster vaccination regimen with Ad26.COV2.S (5×10^{10} vp or 2.5×10^{10} vp or 1×10^{10} vp).

In Cohort 2, participants who previously received primary vaccination with the Pfizer BNT162b2 vaccine will receive a 1-dose booster vaccination with Ad26.COV2.S (5×10^{10} vp or 2.5×10^{10} vp or 1×10^{10} vp).

Information about the disease, correlates of immunity, and safety issues concerning this new pandemic-causing virus are rapidly evolving. Therefore, it is critical to recognize that the approach outlined in this document can change as insights and discussions evolve.

OBJECTIVES AND ENDPOINTS

Objectives	Endpoints
Primary	
Primary Objective 1a: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level. ***If Primary Objective 1a is met, Primary Objective 1b will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 100*(1-2*α)% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 100*(1-2*α)% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3.

Objectives	Endpoints
	AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 1b: To demonstrate the NI of the neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, if feasible. ***If Primary Objective 1b is met, Primary Objectives 1c and 2a will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the leading variant of high consequence or concern*, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the leading variant of high consequence or concern*, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 100*(1-2*α)% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 100*(1-2*α)% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 1c: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level. ***If Primary Objective 1c is met, Primary Objective 1d will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND

Objectives	Endpoints
	• The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 1d: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (1×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 2a: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2 ***If Primary Objective 2a is met, Primary Objective 2b will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain in serum samples of approximately 300 individuals, collected 2 weeks to 2 months after completing 2-dose primary vaccination with Pfizer BNT162b2 (further referred to as Pfizer BNT162b2 external samples). *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster -

Objectives	Endpoints
	Pfizer BNT162b2 primary regimen [external samples]) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/ post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2b: To demonstrate the NI of neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by 2-dose primary vaccination with Pfizer BNT162b2, if feasible ***If Primary Objective 2b is met, Primary Objectives 2c will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) against the leading variant of high consequence or concern* 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) against the leading variant of high consequence or concern* in Pfizer BNT162b2 external samples *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster - Pfizer BNT162b2 primary regimen [external samples]) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2c: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2.

Objectives	Endpoints
by 2-dose primary vaccination with Pfizer BNT162b2 ***If Primary Objective 2c is met, Primary Objective 2d will be tested.***	• Serological response to vaccination and antibody titers (VNA) against the original strain, in Pfizer BNT162b2 external samples. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster-Pfizer BNT162b2 primary regimen [external samples]) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2d: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (1×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2. • Serological response to vaccination and antibody titers (VNA) against the original strain, in Pfizer BNT162b2 external samples . *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster-Pfizer BNT162b2 primary regimen [external samples]) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.

Objectives	Endpoints
Secondary	
To assess the safety and reactogenicity of Ad26.COV2.S administered at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels administered as booster vaccinations in adults.	• Solicited local and systemic adverse events (AEs) for 7 days after booster vaccination. • Unsolicited AEs for 28 days after booster vaccination. • Serious adverse events (SAEs) and adverse events of special interest (AESIs) throughout the study (from booster vaccination until end of the study).
To assess the neutralizing and binding antibody response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, induced by booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, in adults who have previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain, leading variant of high consequence or concern* AND other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination. • Antibodies binding to SARS-CoV-2 relevant variants of concern or individual SARS-CoV-2 proteins (eg, S and/or receptor-binding domain [RBD] proteins from the SARS-CoV-2 variants of concern) by ELISA and/or MSD.
To assess the neutralizing and binding antibody response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, induced by booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, in adults who have previously completed primary vaccination with Pfizer BNT162b.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain, leading variant of high consequence or concern* AND other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination. • Antibodies binding to SARS-CoV-2 relevant variants of concern or individual SARS-CoV-2 proteins (eg, S and/or receptor-binding domain [RBD] proteins from the SARS-CoV-2 variants of concern) by ELISA and/or MSD.
To assess previous or concomitant infection with SARS-CoV-2 at baseline.	Antibodies binding to the SARS-CoV-2 nucleocapsid (N) protein at Day 1 (N-serology).
Exploratory	
To assess the neutralizing antibody responses against the original strain, leading variant of high consequence or concern* and other relevant variants of concern, following Ad26.COV2.S booster vaccination in participants that previously received Ad26.COV2.S or Pfizer BNT122b2 as primary vaccination, compared observationally to antibody responses 14 days post primary vaccination in participants from Study COV3009 (2-dose schedule of Ad26.COV2.S spaced by 56 days).	• Serological response to vaccination and antibody titers measured by VNA against the original strain, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively. • Serological response to vaccination and antibody titers measured by VNA against the leading variant of high consequence or concern*, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary

Objectives	Endpoints
	vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively. • Serological response to vaccination and antibody titers measured by VNA against other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively. • Serological response to vaccination and antibody titers measured by VNA against the original variant, 14 days after completing Ad26.COV2.S primary vaccination as a 2-dose schedule spaced by 56 days (5×10^{10}vp dose level), in a subset of participants from study COV3009.
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp vs those receiving 2.5×10^{10} vp, in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp or 5×10^{10} vp dose level).
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp vs those receiving 1×10^{10} vp, in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (1×10^{10} vp or 5×10^{10} vp dose level).
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Pfizer BNT162b.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10} vp) in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and after booster vaccination at the 5×10^{10} vp dose level in participants primed with Pfizer BNT162b.
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses against the original strain induced by booster vaccination with 2.5×10^{10} vp in adults who previously completed primary vaccination with Pfizer BNT162b.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination at the 5×10^{10} vp dose level in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and booster vaccination at the 2.5×10^{10} vp dose level in participants primed with Pfizer BNT162b.

Objectives	Endpoints
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses against the original strain induced by booster vaccination with 1×10^{10} vp in adults who previously completed primary vaccination with Pfizer BNT162b.	Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination at the 5×10^{10} vp dose level in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and booster vaccination at 1×10^{10} vp dose level in participants primed with Pfizer BNT162b.
To determine if any of the primary objectives that meet a non-inferiority criterion also meet a superiority criterion.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp OR 2.5×10^{10} vp OR 1×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S OR 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination.
To make observational comparisons between the immunologic response to booster vaccination and the primary vaccination regimen, in participants primed with the Pfizer BNT162b regimen (if appropriate serum samples are available for testing).	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp OR 2.5×10^{10} vp OR 1×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after completing 2-dose primary vaccination with Pfizer BNT162b2
To evaluate patient-reported outcomes (PROs) in relation to the presence of SARS-CoV-2 infection and the presence, severity and duration of COVID-19 signs and symptoms in participants who receive booster vaccination.	• Presence, severity and duration of COVID-19 signs and symptoms • Confirmation of SARS-CoV-2 infection by molecular testing • Evaluate all PCR and N ELISA confirmed cases of SARS-CoV-2 infections according to the charter of the Clinical Severity Adjudication Committee.

Objectives	Endpoints
To explore the relative vaccine efficacy (rVE) of Ad26.COV2.S heterologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate, moderate to severe/critical, mild, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) compared to Ad26.COV2.S homologous booster vaccination at the same dose level (5×10^{10}vp, 2.5×10^{10}vp and 1×10^{10}vp).	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate, moderate and severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.
To explore the rVE of different dose levels of Ad26.COV2.S homologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate, moderate to severe/critical, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) as follows: 5×10^{10}vp versus 1×10^{10}vp dose level 2.5×10^{10}vp versus 1×10^{10}vp dose level	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate, moderate and severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.
To explore the rVE of different dose levels of Ad26.COV2.S heterologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate, moderate to severe/critical, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) as follows: 5×10^{10}vp versus 1×10^{10}vp dose level 2.5×10^{10}vp versus 1×10^{10}vp dose level	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate, moderate to severe/critical and severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.

Objectives	Endpoints
If feasible, to investigate the effect of post booster cellular responses, mRNA profiles, neutralizing responses and/or other functional antibody responses on the probability of experiencing a COVID-19 event, moderate and moderate to severe/critically ill COVID-19 disease, or asymptomatic SARS-CoV-2 infection.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the reference strain and/or SARS-CoV-2 variants, by ELISA/MSD and/or other functional antibody assays, 14 days, 6 months and 1 year after homologous or heterologous booster vaccination with Ad26.COV2.S, if sufficient data are available. • Cellular response to vaccination as measured by flow cytometry, ELISPOT and/or transcriptomics, 14 days, 6 months and 1 year after homologous or heterologous booster vaccination with Ad26.COV2.S, if sufficient data are available. • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post booster vaccination in study COV2008, if sufficient data are available. • COVID-19 cases meeting the criteria for "moderate and moderate to severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available. • Asymptomatic SARS-CoV-2 infection, with onset at least 14 days post vaccination in COV2008, if sufficient data are available. • Analysis of gene expression by RNA transcript profiling and correlation with humoral and cellular immune responses.
In a subset, to assess the cellular immune response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, at baseline (Day 1, pre-booster vaccination) and 14 days, 6 months and 1 year after booster vaccination in adults. *This objective will be tested only if PBMC collection is feasible.*	T helper (Th) 1 and Th2 immune responses as assessed by: • Flow cytometry after SARS-CoV-2 S protein peptide stimulation of peripheral blood mononuclear cells (PBMC) and intracellular staining (ICS) including $CD4^+/CD8^+$, interferon gamma (IFNγ), interleukin (IL)2, tumor necrosis factor alpha (TNFα), IL-4, IL-5, IL13, and/or other Th1/Th2 markers. AND/OR • Dual or single IFNγ and IL-4 enzyme-linked immunospot (ELISpot) assay after stimulation of PBMCs with SARS-CoV-2 S protein peptides.
In a subset of participants, to assess the cellular immune response to the original strain, leading variant of high consequence or concern* and other relevant variants of interest, after booster	T helper (Th) 1 and Th2 immune responses as assessed by: • Flow cytometry after SARS-CoV-2 S protein peptide stimulation of peripheral blood

Objectives	Endpoints
vaccination with Ad26.COV2.S in adults who have completed a primary vaccination regimen with Ad26.COV2.S or BNT162b2, at baseline (Day 1, pre-booster vaccination) and 14 days, 6 months and 1 year after booster vaccination. *This objective will be tested only if PBMC collection is feasible.*	mononuclear cells (PBMC) and intracellular staining (ICS) including $CD4^+/CD8^+$, interferon gamma (IFNγ), interleukin (IL)2, tumor necrosis factor alpha (TNFα), IL-4, IL-5, IL13, and/or other Th1/Th2 markers. AND/OR • Dual or single IFNγ and IL-4 enzyme-linked immunospot (ELISpot) assay after stimulation of PBMCs with SARS-CoV-2 S protein peptides.
To further explore the humoral immune response to Ad26.COV2.S booster vaccination in adults.	Exploratory analyses may include the following assays: • SARS-CoV-2 neutralization as assessed by alternative SARS-CoV-2 neutralization assays (VNA). • Adenovirus neutralization. • Functional and molecular antibody characterization including Fc-mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype. • Epitope-specificity characterization of antibodies. • Cytokine profiling: Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma. • Passive transfer: Analysis of immune mediators correlating with protection against experimental SARS-CoV-2 challenge in a suitable animal model.
In a subset, to assess humoral and cellular responses (if feasible) 28 days following the second dose of Pfizer BNT162b2 against the original strain, leading variant of high consequence or concern* and other relevant variant strains for comparison to booster responses in the different regimens and the primary response to Ad26.COV2.S.	• Assay panels similar to those tested for the other groups will be utilized based on availability of serum and cells. Assessments performed with different assays to those utilized for other groups in this study may be utilized.
At one site (BIDMC), in-depth humoral and cellular immunogenicity assessment may be performed on participants enrolled at that site and in a subset of participants enrolled at other sites.	Exploratory analyses may include the following assays: • SARS-CoV-2 neutralization as assessed by alternative SARS-CoV-2 neutralization assays (VNA). • Adenovirus neutralization.

Objectives	Endpoints
	• Functional and molecular antibody characterization including Fc-mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype. • Epitope-specificity characterization of antibodies. • Cytokine profiling: Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma. • Passive transfer: Analysis of immune mediators correlating with protection against experimental SARS-CoV-2 challenge in a suitable animal model. • Analysis of gene expression by RNA transcript profiling
To characterize and establish correlations between mRNA profiling and immunologic responses to the booster vaccinations.	• Analysis of gene expression by RNA transcript profiling and correlation with humoral and cellular immune responses.
To explore markers of inflammation and upregulation of pathways that may be involved in the thrombosis with thrombocytopenia syndrome (TTS) rarely occurring after vaccination with Ad26.COV2.S.	• Descriptive comparison of RNA sequence profiles from participants receiving Ad26.COV2.S vaccination to the RNA profiles of participants receiving other Ad26-based vaccines.
Explore the relationship between humoral and cellular (if feasible) responses in relationship to the time between primary and booster immunization	• In silico modelling of the humoral and cellular immune responses elicited by Ad26.COV2.S as primary regimen and by Ad26.COV2.S as booster, over time.
To assess the vaccine recipient's risk of developing thrombotic events after Ad26.COV2.S administration.	• Platelet count pre-vaccination at Day 1 (all participants) and Day 29 post-vaccination (for participants with a suspected AESI). • Based on the clinical evaluation of the suspected AESI (eg, whether thrombocytopenia is observed with a thrombotic event), a panel of coagulation-related tests may be conducted on the stored pre-vaccination sample (retrospective test), on Day 29 post vaccination, and on the samples obtained as part of the AESI investigation).

***** As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Hypothesis

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively by vaccine group. The neutralizing antibody response 14 days post Ad26.COV2.S booster vs the neutralizing antibody response 28 days post primary vaccination with Ad26.COV2.S will be used to test the following 7 hypotheses:

Cohort 1

1. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.
2. Non-inferiority of neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, if feasible.
3. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.
4. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.

Cohort 2

5. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by a 2-dose primary vaccination with Pfizer BNT162b2.
6. Non-inferiority of neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by a 2-dose primary vaccination with Pfizer BNT162b2, if feasible
7. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by a 2-dose primary vaccination with Pfizer BNT162b2.
8. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by a 2-dose primary vaccination with Pfizer BNT162b2.

* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

OVERALL DESIGN

This randomized, double-blind, parallel, multicenter study will assess the immunogenicity, reactogenicity and safety of a booster dose of Ad26.COV2.S in adults ≥18 years of age, who have previously received primary vaccination with Ad26.COV2.S or BNT162b2.

The primary immunogenicity endpoints are designed to sequentially show non-inferiority of the responses after boosting with various doses of Ad26.COV2.S in volunteers either primed with Ad26.COV2.S at a 5×10^{10} vp dose level, or Pfizer BNT162b2, vs the neutralizing antibody responses to the primary 1-dose regimen of Ad26.COV2.S which has shown efficacy in a prospective randomized Phase 3 efficacy trial. This study will therefore attempt to show which booster regimens will induce neutralizing antibody responses non-inferior to those seen with the Janssen 1-dose regimen of Ad26.COV2.S which has demonstrated efficacy.

In Cohort 1, a target of approximately 770 participants who have received Ad26.COV2.S in study VAC31518COV3001 (ENSEMBLE) will initially be randomized in a 1:1:1 ratio into 3 groups to receive a 1-dose booster vaccination regimen with Ad26.COV2.S until the group 3 is fully enrolled. Thereafter, randomization will continue in a 1:1 ratio in groups 1 and 2 (see the table below). When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 – Group 1 (Primary objectives 1a and 1b) will be performed on available data from the Cohort 1 – Group 1 participants. The other primary objectives will only be tested at the primary analysis.

In Cohort 2, a target of approximately 770 participants who have received primary vaccination with the Pfizer BNT162b2 vaccine will initially be randomized in a 1:1:1 ratio into 3 groups to receive a 1-dose booster vaccination regimen with Ad26.COV2.S until group 6 is fully enrolled. Thereafter, randomization will continue in a 1:1 ratio in groups 4 and 5 (see table below).

Schematic Overview of Study Design and Groups

Cohort	Group	N	Day 1 Vaccination
Cohort 1 (Ad26.COV2.S primary vaccination)	1	~ 330	Ad26.COV2.S at 5×10^{10} vp
	2	~ 330	Ad26.COV2.S at 2.5×10^{10} vp
	3	~ 110	Ad26.COV2.S at 1×10^{10} vp
Cohort 2 (BNT162b2 primary vaccination)	4	~ 330	Ad26.COV2.S at 5×10^{10} vp
	5	~ 330	Ad26.COV2.S at 2.5×10^{10} vp
	6	~ 110	Ad26.COV2.S at 1×10^{10} vp

In Cohort 2 participants, the study vaccine will first be administered to 6 sentinel participants (2 participants per group), enrolled at the same study site, to monitor for unexpected severe adverse reactions. The sentinel participants will be vaccinated at least 1 hour apart and will be closely observed for a minimum of 30 minutes post-vaccination for the development of acute reactions. Each sentinel participant is scheduled to come to the site for a study visit on Day 2 (approximately 24 hours post-vaccination) to collect safety data, which will include solicited AEs (collected through a e-Diary), unsolicited AEs, AESIs, and SAEs. The collected data will be reviewed in a blinded manner by the principal investigator and the sponsor's study responsible physician/scientist. Randomization and vaccination of additional participants will be halted until the review of sentinel data is completed. In the absence of clinically significant findings from the review of 24-hour safety data from the sentinel participants, randomization and vaccination will continue in Cohort 2.

The study duration from screening until the last follow-up visit will be approximately 1 year per participant. The study will consist of a 14-day screening phase, vaccination on Day 1, and follow-up visits up to 1 year after booster vaccination (Target Visit Day 361±21 days).

For each group, safety will be assessed by collection of solicited local (at injection site) and systemic AEs, unsolicited AEs, AESIs, and SAEs. Other safety assessments include clinical laboratory assessments, vital signs measurements (pulse/heart rate, preferably supine systolic and diastolic blood pressure, respiratory rate, and body temperature) and physical examinations.

Active surveillance for COVID-19-like signs and symptoms will occur, including a COVID-19 surveillance (symptom check) through the eCOA. All participants with COVID-19-like signs or symptoms meeting the prespecified criteria for suspected COVID-19 and all participants with at least 1 positive RT-PCR test for SARS-CoV-2 within 5 days of symptom onset, should undertake pre-specified COVID-19 procedures.

The Clinical Severity Adjudication Committee (CSAC) has been established and is tasked with reviewing clinical and study information on cases of COVID-19 that occur in study participants during their time on the study. The CSAC adjudicates the severity of each episode in line with the processes described in the CSAC Charter. The CSAC's assessment of severity is considered the definitive classification of severity for each case. Re-adjudication can occur if new/updated information for a case becomes available. The final adjudication of a case by the CSAC determines its severity for analysis.

The first ~330 randomized participants in each of the cohorts will be assigned to 1 of 4 blood collection subsets (Subsets 1, 2, 3 and 4). Blood samples will be collected for immunogenicity assessments as shown in the tables below. Once the 4 blood collection subsets are enrolled, subsequent participants will not be assigned to a subset. Participants not assigned to a subset ('non-subset' in table below) will have immunogenicity samples taken at Day 1, Day 15, Day 120, Day 181 and Day 361.

Blood Collection Schedule (Whole Blood) for Humoral and Cellular Immunogenicity Assessments

Cohort	Group	Day 1[ab]	Day 2[a,b]	Day 8[ab]	Day 15[ab]	Day 29[a]	Day 71	Day 120	Day 181	Day 361
1	1	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)	Non Subset[c] (N ~380)	All	All
	2	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
	3	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
2	4	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)	Non Subset[c] (N ~380)	All	All
	5	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
	6	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All

a. Selected timepoints include samples for PAXgene analysis.
b. Selected time points include samples for cytokine analysis (Subset 1, 2 and 3 only).
c. Day 120 blood draw only for participants not allocated to a subset

Blood Collection Schedule (PBMC[a]) for Cellular Immunogenicity Assessments

Cohort	Group	Day 1	Day 15	Day 181	Day 361
1	1	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	2	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	3	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
2	4	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	5	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	6	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)

a. Blood collection for PBMC will be performed in a subset of participants in all groups (Subset 4), if PBMC analysis is feasible.

From external sources, the sponsor will obtain serum samples of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used to assess NI of the seropositivity rate induced by a Ad26.COV2.S booster following a primary Pfizer BNT162b2 regimen to the seropositivity post a primary Pfizer BNT162b2 regimen.

Safety issues and concerns will be presented to the Independent Data Monitoring Committee (IDMC) , as appropriate. .

NUMBER OF PARTICIPANTS

Overall, a target of approximately 770 adult participants aged ≥18 years, who have received Ad26.COV2.S in study COV3001 will be enrolled in the study. In addition, a target of approximately 770 participants who have completed primary vaccination with the 2-dose Pfizer BNT162b2 regimen will be enrolled.

DOSAGE AND ADMINISTRATION

Ad26.COV2.S will be supplied at a concentration of 1×10^{11} vp/mL, as suspensions in single-use vials. The study will assess 3 dose levels of Ad26.COV2.S: 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp. Details on study vaccine administration are provided in the Investigational Product Preparation Instructions [IPPI]). Formulation buffer will be supplied as diluent.

MONITORING FOR ASYMPTOMATIC SARS-COV-2 INFECTION

Analysis of antibodies binding to the SARS-CoV-2 N protein will be measured at Day 1 by non-S-protein assays (eg, N protein ELISA and/or SARS-CoV-2 immunoglobulin assay that is dependent on the SARS-CoV-2 N protein).

IMMUNOGENICITY EVALUATIONS

Venous blood samples will be collected for assessment of humoral and cellular immune responses as detailed in the Schedule of Activities.

From external sources, the sponsor will obtain serum samples of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used to assess NI of the seropositivity rate induced by a Ad26.COV2.S booster following a primary Pfizer BNT162b2 regimen to the seropositivity post a primary Pfizer BNT162b2 regimen.

Immunogenicity assays are summarized in the tables below.

Summary of Humoral Immunogenicity Assays

Assay	Purpose
Primary/Secondary endpoints	
SARS CoV 2 neutralization (VNA)	Analysis of neutralizing antibodies to the live SARS CoV 2 original strain and leading variant of high consequence or concern* using a live VNA and/or pseudovirion expressing S protein neutralization assay
Secondary endpoints	
SARS CoV 2 neutralization (VNA)	Analysis of neutralizing antibodies to additional relevant live SARS CoV 2 variants of concern and/or pseudovirion expressing S protein from SARS CoV 2 variants of concern
SARS CoV 2 binding antibodies (ELISA and/or MSD)	Analysis of antibodies binding to SARS CoV 2 or individual SARS CoV 2 proteins (eg, S and RBD proteins from the SARS CoV 2 original strain, leading variant of high consequence or concern* and/or other variants of concern)
Exploratory endpoints	
SARS CoV 2 neutralization (neutralization assay)	Analysis of neutralizing antibodies to the vaccine strain (or other strain), as measured by an alternative neutralization assay (different from the VNA used for the primary and secondary endpoint)
Adenovirus neutralization (neutralization assay)	Analysis of neutralizing antibodies to adenovirus

Assay	Purpose
SARS CoV 2 binding antibodies	Analysis of antibodies binding to SARS CoV 2 or individual SARS CoV2 proteins (eg, S and RBD proteins from the SARS CoV 2 original strain, leading variant of high consequence or concern* and/or other relevant variants of concern) measured by an alternative binding antibody assay (different from the binding antibody assay used in the secondary endpoints)
Functional and molecular antibody characterization	Analysis of antibody characteristics including Fc mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype directed against the original SARS CoV 2 strain, leading variant of high consequence or concern* and/or other variants of concern
Epitope specificity characterization	Analysis of site specificity, epitope mapping
Cytokine profiling	Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma
Passive transfer	Analysis of immune mediators correlating with protection against experimental SARS CoV 2 challenge in a suitable animal model

ELISA enzyme linked immunosorbent assay; Ig immunoglobulin; MSD Meso Scale Discovery; RBD receptor binding domain; S spike; SARS CoV 2 severe acute respiratory syndrome coronavirus 2; VNA virus neutralization assay
* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Summary of Cellular Immunogenicity Assays

Assay	Purpose
Exploratory endpoints	
Flow cytometry (ICS)	Analysis of T cell responses to SARS CoV 2 S protein peptides from the original strain, leading variant of high consequence or concern* and/or other variants of concern by ICS including $CD4^+/CD8^+$, IFNγ, IL 2, TNFα, IL 4, IL 5, IL 13, and/or other Th1/Th2 markers (if feasible)
ELISpot	IFNγ and IL 4 responses to SARS CoV 2 S protein peptides by PBMCs (if feasible), based on single or dual ELISpot
Gene expression analysis	Analysis of gene expression by RNA transcript profiling and/or analysis of protein translates, in cells or whole blood stimulated with SARS CoV 2S protein peptides or in unstimulated cells or whole blood (ex vivo)

ELISpot enzyme linked immunospot (assay); ICS intracellular cytokine staining; IFNγ interferon gamma; IL interleukin; PBMC peripheral blood mononuclear cell; S spike; SARS CoV 2 severe acute respiratory syndrome coronavirus 2; Th T helper; TNFα tumor necrosis factor alpha
* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the start of sample analysis

SAFETY EVALUATIONS

After booster vaccination, participants will remain under observation at the study site for at least 30 minutes to monitor for acute reactions and solicited events. Participants will be asked to note in the e-Diary occurrences of injection site pain/tenderness, erythema and swelling at the study vaccine injection site daily for 7 days post-vaccination (day of vaccination and for the subsequent 7 days). Participants will be instructed on how to record daily temperature using a thermometer provided for home use.

Participants should record the temperature in the e-Diary in the evening of the day of vaccination, and then daily for the next 7 days approximately at the same time each day. Participants will also be instructed on how to note signs and symptoms in the e-Diary on a daily basis for 7 days post-vaccination (day of vaccination and for the subsequent 7 days), for the following events: fatigue, headache, nausea, and myalgia.

Adverse events and special reporting situations, whether serious or non-serious, that are related to study procedures or that are related to non-investigational sponsor products will be reported from the time a signed and dated informed consent form (ICF) is obtained until the end of the study/early withdrawal. All other unsolicited AEs will be reported from the time of vaccination until 28 days post-vaccination. All AESIs, SAEs, and AEs leading to discontinuation from the study (regardless of the causal relationship) are to be reported from the moment of vaccination until completion of the participant's last study-related procedure.

Thrombosis with thrombocytopenia syndrome (TTS) is considered to be an AESI (Adverse Event of Special Interest). Suspected AESIs (thrombotic events and thrombocytopenia [defined as platelet count below 150,000/μL[a]]) will be reported from the moment of vaccination until the end of the study/early withdrawal.

An AESI Adjudication Committee with appropriate expertise will be established to evaluate each suspected AESI and determine whether it meets the definition of TTS.

Any participants who receive a Covid-19 booster vaccine off study will remain on study (continuing with all scheduled procedures and follow-up) but will be excluded from the formal hypothesis testing of the primary objectives.

STATISTICAL METHODS

Sample Size Determination

The following assumptions were made in the sample size determinations:

For the non-inferiority comparisons based on seroresponse rate:

- Under the null hypothesis, a 90% responder rate on Day 15 post -booster and a 90% responder rate on Day 29 post primary vaccination
- Non-inferiority margin of -10% for the difference between 14 days post-booster and 28 days post primary vaccination responder rate on Day 29
- Farrington & Manning Likelihood Score Test used in the non-inferiority testing of the difference between the two responder rates

For the non-inferiority comparisons based on GMT:

- Log transformed (log_{10} scale) VNA data are normally distributed
- A standard deviation (SD) of VNA = 0.56 (log_{10} scale)
- Under the null hypothesis, a GMT ratio (Day 15 post-booster/Day 29 post primary regimen) of 1.0
- Non-inferiority margin = $log_{10}(2/3)$ = -0.176 for the GMT ratio testing
- No correlation between immune responses 28 days post-dose 1 and 14 days post-booster

In addition, a drop-out rate approximately 10% is assumed.

With the above assumptions, the required sample size to achieve approximately 95% power to demonstrate non-inferiority, at alpha 0.025 (1-sided), of primary hypothesis 1a is N=297 (N=330 adjusted for the 10% dropout). With this sample size, the power to demonstrate non-inferiority of response rate is 98% and the power to demonstrate non-inferiority of the geometric mean ratio (GMR) is 97%, resulting in a combined power of approximately 95%.

Similarly, the required sample size to achieve 90% power to demonstrate the non-inferiority, at alpha 0.0125 (1-sided), of the remaining primary hypotheses is N=297 (N=330 adjusted for the 10% dropout). With this sample size, the power to demonstrate non-inferiority of response rate is 95% and the power to

[a] Updated Proposed Brighton Collaboration process for developing a standard case definition for study of new clinical syndrome X, as applied to Thrombosis with Thrombocytopenia Syndrome (TTS). 18 May 2021. https://brightoncollaboration.us/wp-content/uploads/2021/05/TTS-Interim-Case-Definition-v10.16.3-May-23-2021.pdf .Accessed: 02 September 2021.

demonstrate non-inferiority of the geometric mean ratio (GMR) is 94%, resulting in a combined power of approximately 90%.

When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 – Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort – Group 1 participants. If the interim analysis is conducted, an O'Brien-Fleming adjustment will be used whereby the type I error for the non-inferiority test at the interim analysis is 0.0003 (one-sided) and at the final analysis 0.0249. Because an O'Brien-Fleming adjustment is used, the total sample size is not increased for this interim analysis.

Populations for Analysis Sets

For purposes of analysis, the following populations are defined:

FAS: The full analysis set will include all participants with a documented study vaccine administration (Ad26.COV2.S). Analyses of safety and reactogenicity will be performed on the FAS.

PPI: The per protocol immunogenicity population will include all vaccinated participants for whom post-baseline immunogenicity data are available excluding participants with major protocol deviations expected to impact the immunogenicity outcomes. In addition, samples obtained after natural SARS-CoV-2 infection (if applicable) will be excluded from the analysis.

NI: The NI analysis set will include all PPI participants who are SARS-COV-2 seronegative at baseline (based on the serological test for SARS CoV-2-specific nucleocapsid antibodies [N serology]). The NI hypothesis tests, as well as descriptive analysis of secondary endpoints for other VOCs, will be performed on the NI analysis set.

PPE: The per protocol for efficacy analysis set will include all vaccinated participants who are SARS-CoV-2 seronegative at baseline and who have no major protocol deviations expected to impact the efficacy outcomes.

Pfizer BNT162b2 external samples: serum samples from external sources of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used for NI assessments for Cohort 2.

Primary Endpoints

Immunogenicity Endpoints

For NI assessments of primary objectives 1a and 1b (evaluated at the interim or primary analysis), the Ad26.COV2.S booster vaccine at the 5×10^{10} vp dose level against the original strain (objective 1a) and leading variant of concern (objective 1b) at 14 days post booster as compared to the Ad26.COV2.S vaccine against the original strain (objective 1a) and leading variant of concern (objective 1b) at 28 days post-vaccination, two NI hypothesis tests will be performed:

1. Statistical NI of responder rate, with a margin of -10%: the difference in responder rate between post booster minus post prime will be estimated, with a 100*(1-2*α)% CI. Non-inferiority for responder rate will be demonstrated if the 2-sided 100*(1-2*α)% CI lies entirely above -10%.
2. Statistical NI of GMTs, with a margin of 1.5-fold: the ratio of the GMTs (post booster GMT divided by post prime GMT, i.e; the Geometric Mean Ratio [GMR]) will be estimated, with the corresponding 100*(1-2*α)% CI. Non-inferiority will be demonstrated if the 2-sided 100*(1-2*α)% CI of the estimated GMT ratio lies entirely above 2/3.

3. In addition to the above, an estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.

For objective 1a and 1b, at the interim analysis (if conducted), an O'Brien-Fleming adjustment will be used, α=0.0003 and a 99.4% CI will be calculated. At the primary analysis, α=0.0249 and a 95.02% CI will be calculated, if the interim analysis is conducted. Otherwise, α=0.025 and a 95% CI will be calculated.

Similar non-inferiority criteria will be used for the remaining primary objectives 1c, 1d, 2a, 2b, 2c and 2d (evaluated only at the primary analysis), except that 97.5% CI will be used for both the responder rate and GMT hypothesis testing. A hierarchical testing strategy will be applied.

For primary objectives 1a, 1b, 1c and 1d, formal NI testing will be conducted as a "within-subjects" analysis, in which participants' VNA data are considered matched pairs across the two time points. The NI analysis on responder rate will be based on the Agresti-Min (Agresti 2005) method to estimate the difference in proportions and its CI. This method was chosen because of its well-behaved coverage probability (CP) properties compared to other methods for the analysis of matched pairs data (Reed 2009). Coverage probability is generally used to evaluate $(1 - \alpha)$ CIs where α is the Type I error rate. The NI analysis on the GMR will use a paired t-test to estimate the difference in means and its CI on $\log_{10}$ transformed data. The estimated difference and its CI will be back transformed to yield the GMT ratio (GMR) and its CI.

As a sensitivity analysis, 28 days post-dose 1 VNA data may be pooled across the three study arms and compared to the 14 days post booster VNA data in the study arm of interest. This sensitivity analysis will use appropriate statistical models, such as linear mixed models for the GMR and Generalized Estimating Equations (GEE) models for the difference in responder rates.

For primary objectives 2a, 2b, 2c and 2d relating to Cohort 2, formal NI testing will be conducted by comparing VNA data from external samples, collected 2 weeks to 2 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the 14 days post booster VNA data in the study arm of interest (groups 4, 5 or 6).

Secondary Endpoints

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively by vaccine group. All safety analyses will be made on the FAS. The immunogenicity analyses will be performed on the PPI and FAS population.

Descriptive statistics (geometric mean and CIs, or median and interquartile range Q1-Q3, as appropriate) will be calculated for continuous immunologic parameters. Several definitions of serological response will be applied as applicable (GMC [S-ELISA], GMT [VNA]). Graphical representations of immunologic parameters will be made as applicable. Frequency tabulations will be calculated for discrete (qualitative) immunologic parameters, as applicable.

In addition, the ratio between neutralizing and binding antibodies as determined by VNA and S-ELISA, respectively, will be calculated together with CIs.

Planned Analysis

When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 – Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort 1 – Group 1 participants. The other primary objectives will only be tested at the primary analysis.

The primary analysis of safety and immunogenicity in Cohort 1 or Cohort 2 will be performed when all evaluable participants in the respective cohort have completed the visit that takes place 28 days after study

vaccination or discontinued earlier. The analysis will include immunogenicity data (VNA, N-serology, and S-ELISA and/or MSD [if available at the time of analysis]) for all evaluable participants through Day 15 and all available safety data. The sponsor will be unblinded at the time of this primary analysis, with the exception of specific sponsor personnel (see below). If the primary analysis of Cohort 1 is performed before the primary analysis of Cohort 2, then the sponsor will be unblinded only for Cohort 1 at the time of the Cohort 1 primary analysis, and unblinded for only Cohort 2 at the time of the primary analysis of Cohort 2.

For the primary analysis, and for any analyses after the primary analysis on the same cohort(s), unblinded data at the participant level will be available to sponsor personnel including statistical programming, statistics, clinical and clinical immunology personnel involved in the analysis, and the sponsor committee involved in making future decisions for the program. Sponsor personnel directly involved in data collection, data management, and safety monitoring will not have access to unblinded data at the participant level until the study end. Sites and study participants will not have access to unblinded data at the participant level until the end of study, with two exceptions. Firstly, the investigator may, in an emergency, determine the identity of the intervention. Secondly, the sponsor may unblind the study at selected sites after all participants still on study at those sites have reached the 6-month post booster timepoint, to generate a list of participants who may be eligible for enrollment into the sponsor-supported VAC31518COV2015 study. Group level data may be shared with investigators or other blinded clinical staff, as needed, but every effort will be made to preserve the blinding to the individual participant allocation until the end of study.

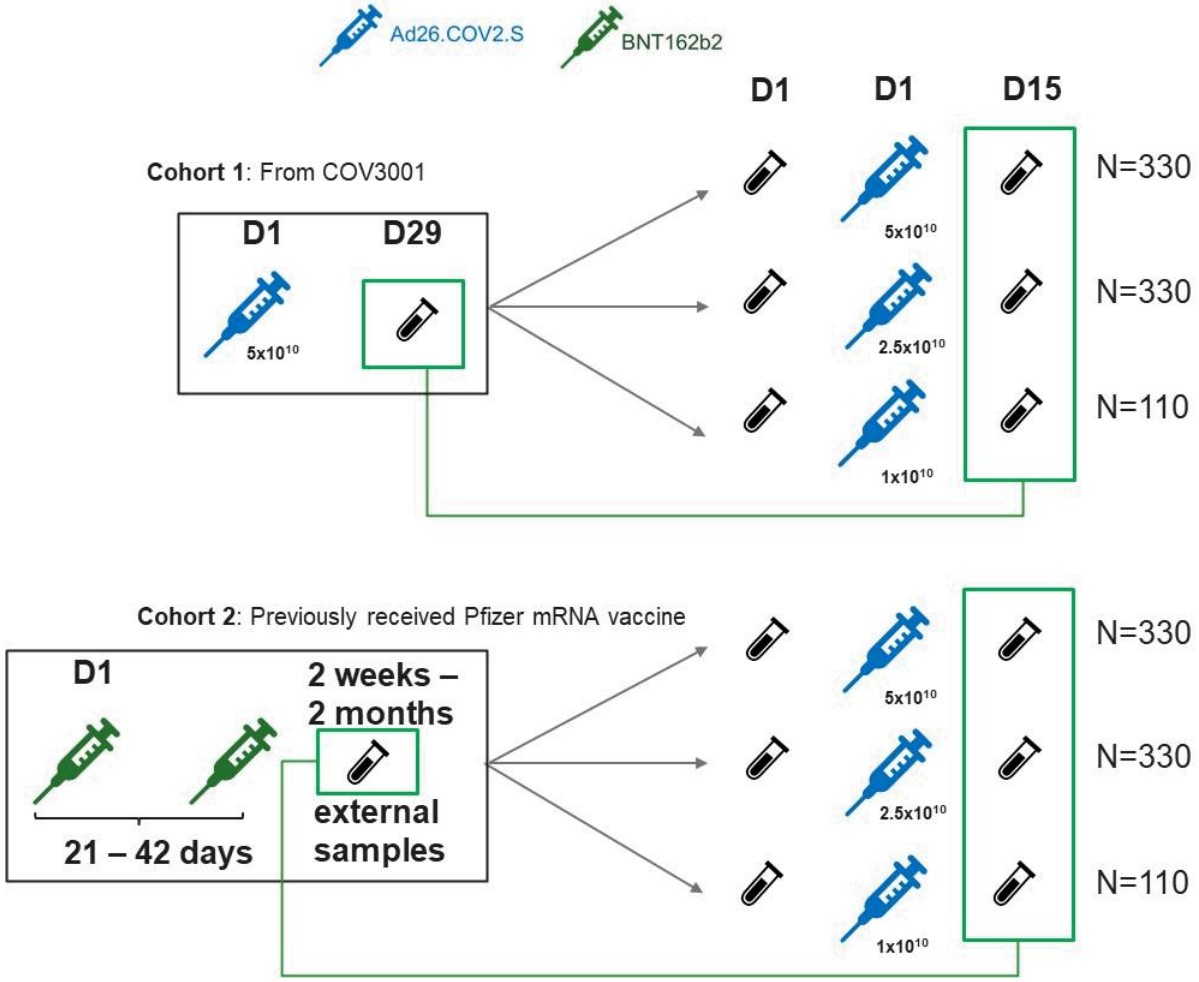
The final analysis in Cohort 1 or Cohort 2 will be performed when all included participants in the respective cohort have completed their last visit or discontinued earlier

There are no planned formal assessments of efficacy in this study. However, data on molecularly confirmed SARS-CoV-2 infections, N protein binding antibodies and COVID-19-like signs and symptoms will be reported after the primary analysis, and relative efficacy may be estimated as permitted by the available data.

After the Primary Analysis, additional Interim Analyses may be performed by the sponsor as required.

1.2. Schema

Figure 1: Schematic Overview of the Study



D= Day, N= number of participants in each vaccine group, NI = non-inferiority

1.3. Schedule of Activities

1.3.1. All Participants

Phase	Screening[a]		Follow-up[b]									
Clinic Visit #	1	2	3[w]	4[u]	5	6[v]	7[w]	7A[bb]	8	9[aa]	10	Exit[x]
Visit Timing		Vac	Vac +1d	Vac + 7 d	Vac + 14 d	Vac + 28 d	Vac + 70 d	Vac + 119 d	Vac + 180 d	Vac + 270	Vac + 360 d	
Visit Day ±Window	-14 to 1	1	2	8±2[c]	15 -3/+7	29 -3/+10	71±14	120±14	181±21	271±21[aa]	361±21	
Visit Type	Screening	Vaccine	Safety	Safety and immuno	Safety and Immuno	Safety and immuno/Safety phone call	Safety and immuno	Safety and immuno	Safety and immuno	Safety phone call[aa]	Safety and immuno	Early Exit
Written informed consent[d]	●											
Inclusion/exclusion criteria	●	●[1]										
Demographics	●											
Medical history/prestudy meds	●											
Physical examination[e]	●											
Vital signs[f] incl. body temperature	●	●[2]	● Subset 3	● Subset 1	● All	● Subset 2	● Subset 3	● No Subset	● All		● All	●
Pregnancy test (for participants of child bearing potential only)	●											
Randomization		●[1]										
Prevaccination check[g]		●[1]										
Clinical laboratory blood sample (whole blood)[i]		●[1]			● All							
Serological test for SARS-CoV 2 specific nucleocapsid antibodies (based on N serology)		●[1]			● All	● Subset 2	● Subset 3	● No Subset	● All		● All	● All
Humoral immunity (serum), blood draw		●[1] All		● Subset 1	● All	● Subset 2	● Subset 3	● No Subset	● All		● All	●[3]
Cellular immunity (PBMC), blood draw[j]		●[1] Subset 4			● Subset 4				● Subset 4		● Subset 4	
Blood draw for exploratory research[z]		●[1]			●	●		●	●		●	●[3]
RNA Sequencing (whole blood, PAXgene® tubes)		●[1] All	● Subset 3	● Subset 1	● All	● Subset 2						
Cytokines, blood draw		●[1] Subset 1,2&3	● Subset 3	● Subset 1	● Subset 2							
Nasal sample collection for SARS CoV 2 testing[k]		●[1]										
Vaccination		●										
30 minute post vaccination observation[l]		●										

Phase	Screening[a]		Follow-up[b]									
Clinic Visit #	1	2	3[w]	4[u]	5	6[v]	7[w]	7A[bb]	8	9[aa]	10	Exit[x]
Visit Timing		Vac	Vac +1d	Vac + 7 d	Vac + 14 d	Vac + 28 d	Vac + 70 d	Vac + 119 d	Vac + 180 d	Vac + 270	Vac + 360 d	
Visit Day ±Window	-14 to 1	1	2	8±2[c]	15 -3/+7	29 -3/+10	71±14	120±14	181±21	271±21[aa]	361±21	
COVID 19 like Symptom surveillance booklet and nasal swab kit and saliva samples training and distribution[o]		●										
Solicited AE recording (ediaries)		-------	*Continuous-*	------								●[4]
Unsolicited AE recording[m]		*---------- -Continuous through +28 d -----*										●[5]
SAE/AESI recording[n]		-------	-------	------	*--- Continuous-- - -- --*		-------	-------	-------	-------	------	----
Concomitant therapies[q]		-------	-------	------	*---- Continuous- - -- --*		-------	-------	-------	-------	------	----
COVID 19 signs and symptoms surveillance[p]			-------	------	*----- Continuous- - -- --*		-------	-------	At least twice a month			
Symptoms of Infection with Coronavirus 19 (SIC), including body temperature measured by the participant (ePROs to be completed by the participant in the eCOA)[p]		●[1]										
Distribution of pulse oximeter[s]		●[1]										
eCOA training/retraining and set up[h]		●[1]										
MA COV form distribution[y]		●[1]										
Participant e diary distribution[r]		●[1]										
Participant e diary review[t]				● Subset 1	● All							

●[1] pre vaccination; ●[2] pre and post vaccination; ●[3] blood samples for immunogenicity will only be taken if the early exit visit is at least 10 days after the previous immunogenicity blood draw; ●[4] if within 7 days of vaccination; ●[5] if within 28 days of vaccination.

a. Screening for COV2008 will be performed within 14 days prior to booster vaccination or on the day of booster vaccination. 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 booster vaccination.

b. If a participant shows COVID-19 like symptoms, the participant should contact the site for guidance and must follow their local country and site level recommendations for COVID-19.

c. If a participant comes in early for Visit 4, ie, 1 or 2 days prior to the Visit Day per the allowed visit window, a subsequent phone call will be made at the end of the diary period to collect diary information recorded between the actual visit and the end of the diary period. The e-Diary will be returned by the participant at the next visit.

d. The ICF(s) must be signed before any study-related activity.

e. A history-directed physical examination, including height and body weight, will be carried out at screening. At other visits, an abbreviated, symptom-directed examination will be performed if determined necessary by the investigator.

f. Pulse/heart rate, systolic and diastolic blood pressure, respiratory rate, and body temperature. Blood pressure measurements should be taken in the supine position (preferably) and preceded by at least 5 minutes of rest. Vital signs measurements are recommended before blood sampling. Body temperatures will be measured preferably via the oral route.
g. Investigator must check for acute illness or body temperature ≥38.0°C/100.4°F at the time of vaccination. If any of these events occur at the scheduled time for vaccination, the vaccination can be rescheduled as long as this is in agreement with the allowed window. If the vaccination visit cannot be rescheduled within the allowed window or the contraindications to vaccination persist, the sponsor should be contacted for further guidance. The investigator should also check if any other reasons have been met and would prevent study vaccination.
h. eCOA set up and training to be provided for Cohort 2 participants. eCOA set up and training will have been conducted for all COV3001 participants. If required, retraining will be provided for Cohort 1 COV2008 participants. All COV2008 participants will complete the eCOA using an application on their own eDevice (smartphone or tablet) if their device is compatible with the application or using the web portal. All eCOA assessments should be conducted/completed before any tests, procedures, or other consultations to prevent influencing participant responses. If a participant is unable to complete the eCOA, a study staff member or the participant's caregiver can collect information on the participant's behalf as detailed in Section 8.1.2.
i. Whole blood samples will be used for a platelet count (as part of a complete blood count if applicable) in a local laboratory or substitute for local laboratory, depending on local feasibility towards turnaround time of sample processing. Results are to be reported in the eCRF. Serum and plasma samples will be derived from the whole blood sample and stored for potential future coagulation-related testing in a central laboratory if the participant experiences a suspected AESI (see Appendix 2).
j. Collection of blood samples for PBMC analysis will be performed in a subset of participants in all groups (Subset 4), if PBMC analysis is feasible.
k. Nasal swabs to be batch tested centrally (result is NOT required for randomization).
l. Participants will be closely observed for a minimum of 30 minutes post-vaccination. Any solicited local (at injection site) and systemic AEs, unsolicited AEs, AESIs, SAEs, concomitant medications, and vital signs will be documented by study-site personnel following this observation period and participants will be allowed to leave the study site after it is documented that the 30-minutes post-vaccination observation period is complete.
m. AEs and special reporting situations that are related to study procedures or that are related to non-investigational sponsor products will be reported from the time a signed and dated ICF is obtained until the end of the study/early withdrawal. All other unsolicited AEs and special reporting situations will be reported from the time of vaccination until 28 days post-vaccination.
n. All SAEs related to study procedures or non-investigational sponsor products will be reported for all participants from the time a signed and dated ICF is obtained until the end of the study/early withdrawal. All other SAEs are to be reported for all participants from the moment of vaccination until completion of the participant's last study-related procedure. AEs leading to study discontinuation are to be reported from the moment of vaccination until completion of the participant's last study-related procedure. Suspected AESIs are to be reported from the moment of vaccination until completion of the participant's last study-related procedure (see Section 8.4.1).
o. Cohort 1 participants will have received COVID-19-like Symptom surveillance booklet, nasal swab kit and saliva sample training for COV3001. If required, replacement kits and/or retraining will be provided prior to booster vaccination. Booklet, nasal swab kit and training to be provided to Cohort 2 participants.
p. The SIC questionnaire asks the participant if he/she had any of the prespecified signs or symptoms (see Appendix 5) during the past 24 hours (including highest temperature in the last 24 hours), and (when applicable) to rate the severity. If a participant develops COVID-19-like signs and symptoms, refer to Section 1.3.3 and Section 8.1.2.
q. Concomitant therapies such as analgesic/antipyretic medications and non-steroidal anti-inflammatory drugs, corticosteroids, antihistamines, and vaccinations must be recorded from the moment of vaccination until 28 days after administration of study vaccine. All other concomitant therapies should also be recorded if administered in conjunction with a confirmed COVID-19 case or with new or worsening AEs reported per protocol requirements outlined in Section 8.4.1.
r. The e-Diary, a ruler and thermometer to be distributed to Cohort 2 participants. Materials will have been distributed to Cohort 1 participants for COV3001, replacement materials and/or retraining will be provided if required.

s. Pulse oximeters to be provided to Cohort 2 participants at their baseline visit to measure blood oxygen saturation and pulse rate during a COVID-19 episode (see Section 1.3.3). All Cohort 1 participants will have been provided a pulse oximeter at their baseline COV3001 visit, any misplaced/malfunctioning oximeters will be replaced prior to booster vaccination on study COV2008.
t. If an event is still ongoing 7 days after the vaccination, the participant should continue to collect information in the diary until resolution.
u. Visit for participants in immunogenicity subset 1 only.
v. Visit for participants in immunogenicity subset 2 only and a phone call will be made to the participants not in immunogenicity subset 2 to confirm the collection of unsolicited AEs.
w. Visit for participants in immunogenicity subset 3 only. Window for Visit 3 is 24 hours post vaccination for sentinel participants and 24 to 72 hours post vaccination for non-sentinel participants.
x. For those participants who are unable to continue participation in the study up to Visit 7, but for whom consent is not withdrawn, who are not lost to follow-up, or who have not died, an early exit visit will be conducted as soon as possible. Participants who wish to withdraw consent from participation in the study will be offered an optional visit for safety follow-up. This includes the safety assessments of the early exit visit (no blood sampling for immunogenicity).
y. MA-COV form to be provided to Cohort 2 participants at the vaccination visit. The MA-COV form (Appendix 7) will have been provided to all Cohort 1 participants at the COV3001vaccination visit. If needed, a new form should be provided prior to booster vaccination on study COV2008. The MA-COV form should be completed by the medical care provider or the study site personnel during medical visits for COVID-19 or COVID-19 complications.
z. Only applicable to participants enrolled at the BIDMC site.
aa. The site staff will phone the participants to ensure retention and to check on the occurrence of SAEs, AESIs or MAAEs.
bb. Only for participants not assigned to a blood drawing subset, and participants enrolled at BIDMC.

AE = adverse event; AESI = adverse event of special interest; COVID-19 = coronavirus disease-2019; d = day(s); eCOA= electronic clinical outcome assessment; eCRF = electronic case report form, ICF = informed consent form; PBMC = peripheral blood mononuclear cell; SAE = serious adverse event; SARS-CoV-2 = severe acute respiratory syndrome coronavirus-2; SIC = Symptoms of Infection with Coronavirus-1; vac = vaccination.

1.3.2. Participants with a Suspected AESI

The medical management of thrombotic events with thrombocytopenia is different from the management of isolated thromboembolic diseases. Study site personnel and/or treating physicians should follow available guidelines for treatment of thrombotic thrombocytopenia (eg, from the American Society of Hematology 2021; British Society of Haematology - Expert Haematology Panel 2021, and the CDC 2021d). The use of heparin may be harmful and alternative treatments may be needed. Consultation with a hematologist is strongly recommended. Management of the participant should not be delayed by decision-making of the AESI Adjudication Committee. In the event of a suspected thrombotic event, thrombocytopenia, or TTS, laboratory assessments (to be performed locally) are required to facilitate diagnosis and determine treatment options, including but not limited to platelet count and anti-PF4 tests.

Additional blood samples should be collected for central laboratory testing as detailed below. However, results of central laboratory testing may not be available to guide immediate treatment decisions.

Timing relative to onset of suspected AESI	**AESI Day 1[a]**	**AESI Day 29[b]**
Visit window		**±7 d**
Site to report AESI[c]	●	
Clinical lab blood sample (whole blood)[d]	●	●
TTS AESI form[e]	*- - - - Continuous- - - - -*	
Concomitant therapies[f]	●	●

a. Day 1 refers to first awareness of the event, which might be later than the date of onset. Every effort should be made to report as much information as possible about the event to the sponsor in a reasonable timeframe. The investigator should contact the sponsor for input on the feasibility of collecting blood samples, including the need for additional samples based on the nature of the event.
b. Day 29 is to be calculated relative to the actual day of onset of the event. If the event is not resolved on Day 29, subsequent follow-up assessments can be performed at unscheduled visits as needed until resolution of the event. If the event is reported to the investigator more than 28 days after the onset of the event, the AESI Day 29 visit becomes redundant and does not need to be performed.
c. Suspected AESIs must be reported to the sponsor within 24 hours of awareness, irrespective of seriousness (ie, serious and non-serious AEs) or causality assessment, using the SAE form (see Section 8.4.7).
d. Whole blood samples will be used for a platelet count (as part of a complete blood count, if applicable) in a local laboratory or substitute for local laboratory, depending on local feasibility towards turnaround time of sample processing. Results are to be reported in the eCRF. Serum and plasma samples will be derived from the whole blood sample for coagulation-related testing in a central laboratory (see Appendix 2). For the follow-up visit, the volume of blood to be collected may vary depending on the clinical evaluation of the case.
e. Medical information on local case management will be collected. Upon becoming aware of the suspected AESI, study site personnel should provide information on an ongoing basis. See Section 8.4.7 and Appendix 11 for further details.
f. Refer to Section 6.7 for collection and recording of concomitant therapies associated with a suspected AESI.

AE =adverse event; AESI = adverse event of special interest; CDC = Centers for Disease Control and Prevention; PF4 = platelet factor 4; TTS = thrombosis with thrombocytopenia syndrome

1.3.3. Participants With COVID-19-like Signs and Symptoms

Timing relative to onset of signs and symptoms	COVID-19 Day 1-2	COVID-19 Day 3-5[a]		2-day cycle to be repeated[b,c,d,e]		COVID-19 Day 29 (±7 d)[f,g]
		Part 1	Part 2[b]	1st day of cycle	2nd day of cycle	
Location	**Home[h]**	**Site or Home[i,j]**	**Site or Home[i,j]**	**Home[j]**	**Home[j]**	**Site or Home[i,j]**
Participant to contact study site with any health concerns/participant notifies the site of becoming aware of a positive RT-PCR test	●					
Site to contact participant if COVID-19 signs or symptoms are recorded in eCOA	●					
Confirmation of suspected COVID-19 using prespecified criteria	●[k]	●[l]				
Nasal swab sample (collected by the participant at home)[m]	●[n]			●		
Nasal swab sample (collected by qualified study staff)		●[o]				
Saliva sample (collected by the participant)[p]			●		●	
Humoral immunity (serum)			●			●[q]
Biomarker RNAseq blood sample (PAXgene tubes, whole blood)[r]			●			●
In case of signs and symptoms: Symptoms of Infection with Coronavirus-19 (SIC), including highest body temperature over the last 24 hours measured by the participant[s] (ePROs to be completed by the participant in the eCOA)	*Daily*					●[t]
In case of no signs or symptoms: (Suspected) COVID-19 surveillance (symptom check)	*At least twice a month*					●
Vital signs[u]		●				●
Targeted physical examination		●				●
Pulse oximetry by site staff		●				●
Pulse oximetry by the participant (ePRO to be completed by the participant in the eCOA)[v]	●[n]	*3 times a day*				
Medical history (including recent flu or pneumococcal vaccination) and description of COVID-19 episode (collected by interview with the participant)			●			●
MRU questionnaire (collected by interview with the participant)[w]			●			●
Capture medical information from medical visits for COVID-19 or COVID-19 complications (MA-COV form)[x]	*Continuous*					
Concomitant therapies associated with COVID-19	*Continuous*					
Study-site personnel to contact participant	*Weekly or more frequently*					

a. The visit at COVID-19 Day 3-5 should be scheduled 2 to 4 days after symptoms onset/positive RT-PCR test from outside the study.
b. Only applicable for participants that meet the prespecified criteria for suspected COVID-19 (Section 8.1.1) on COVID-19 Day 1-2 and COVID-19 Day 3-5 or who have a positive test result for SARS-CoV-2 on COVID-19 Days 1-2 or 3-5 visits.

c. Participants should be encouraged by the site to collect nasal swabs and saliva samples as indicated in the Schedule of Activities. If the participant is unable or unwilling to collect all samples as requested, the participant should still complete the other COVID-19 assessments, including the visit at COVID-19 Day 29.
d. As soon as it is confirmed that both nasal swabs (collected on COVID-19 Day 1-2 and COVID-19 Day 3-5) are negative for SARS-CoV-2, the participant will not undertake any further COVID-19 procedures and will fall back to the default Schedule of Activities, until the end of the study/early withdrawal.
e. Participants should undertake the COVID-19 procedures until 14 days after symptom onset (COVID-19 Day 15) or until resolution of the COVID-19 episode, whichever comes last. Resolution of a COVID-19 episode is defined as having 2 consecutive SARS-CoV-2 negative nasal swabs and 2 consecutive days with no COVID-19-related signs or symptoms. Once past COVID-19 Day 15, participants should stop the collection of nasal swabs and saliva samples as soon as 2 consecutive nasal samples are SARS-CoV-2 negative, but (if still symptomatic at that time) should continue completing the ePROs (including SIC, body temperature, and pulse oximetry) in the eCOA until 2 consecutive days with no COVID-19-related signs or symptoms (Section 8.1.1).
f. Only applicable for participants that have at least 1 SARS-CoV-2 positive nasal swab collected on COVID-19 Day 1-2 or Day 3-5. COVID-19 Day 29 visit should still be performed even if the nasal swabs results are still pending.
g. The visit on COVID-19 Day 29 can be combined with a regular study visit if within the applicable visit windows.
h. The COVID-19 Day 1-2 nasal swab can be collected at the study site (or hospital or other location, if needed), if preferred by the participant.
i. All COVID-19 Day 3-5 and Day 29 assessments may be performed by a trained HCP at the participant's home, if allowed per local and/or institutional regulations.
j. If a participant has a positive test result for SARS-CoV-2 infection and/or depending on the medical status of the participant, the participant may be requested to remain at home and not visit the study site. If necessary, study-site personnel or a trained HCP will visit the participant at home (or at the hospital or other location, if needed), if allowed by local regulations. Under these circumstances, the participant will be contacted by the site at least once per week and the participant's medical care provider will be notified.
k. In case of COVID-19 like symptoms, based on the information collected through the SIC, the site will reach out to the participant at the latest on COVID-19 Day 2 (the day after the day of symptom onset) to assess whether the reported signs and symptoms qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1). As several of the prespecified criteria for suspected COVID-19 overlap with vaccine-related reactogenicity, investigators' clinical judgement is required to exclude vaccine-related events. In case the participant would actively reach out to the site already on COVID-19 Day 1, the site should already make a first assessment on COVID-19 Day 1 to check whether the reported signs and symptoms qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1).
l. In case of COVID-19 like symptoms, the site will interview the participant to assess whether the reported signs and symptoms still qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1).
m. A nasal swab should be collected from the participant at home (using available material for home swabs provided by the study staff) as soon as the prespecified criteria for suspected COVID-19 are met and, in case of COVID-19 like symptoms, preferably on the day of symptom onset or the day thereafter (COVID-19 Day 1-2). The sample collected on COVID-19 Day 1-2 should be transferred to the study site, as arranged by the study site, as soon as possible after collection, preferably within 24 hours. Nasal swabs should also be collected once every 2 days until 14 days after symptoms onset (COVID-19 Day 15) or until resolution of the COVID-19 episode, whichever comes last. These samples should be transferred to the study site, as arranged by the study site, within 3 days after collection. Details are provided in the laboratory manual. If the participant requires assistance, a trained HCP can help the participant to collect the nasal swabs.
If 2 consecutive nasal swabs negative for SARS-CoV-2 are not available due to operational reasons (eg, delays in results availability), participants may cease collection of nasal swabs and saliva samples after COVID-19 Day 29, provided they have 2 consecutive days with no COVID-19-related signs and symptoms. In these cases, participants may be asked to resume sample collection if nasal sample results—once available—do not present with 2 consecutive negative swabs for SARS-CoV-2.
n. The nasal swab should be collected and pulse oximetry should be started as soon as possible after it has been confirmed that the prespecified criteria for suspected COVID-19 (Section 8.1.1) are met.
o. All nasal swabs will also be tested by a local laboratory for case management.

p. Saliva samples should be collected from the participant (using recipients provided by the study staff). The samples should be transferred to the study site, as arranged by the study site, within 3 days after collection. Details are provided in the laboratory manual. If the participant requires assistance, a trained HCP can help the participant to collect the saliva samples.
q. Blood sample for humoral immunity also includes sample for sero-confirmation of SARS-CoV-2 infection (antibody).
r. Blood sample for exploration of biomarkers correlating with SARS-CoV-2 infection and COVID-19 severity.
s. Participants should complete the (suspected) COVID-19 surveillance (symptom check). In case of COVID-19 like signs and symptoms, participants should be encouraged by the site to complete the SIC (Appendix 5) daily, preferably in the evening around the same time each day, starting on the first day they experience symptoms. Sites should remind the participant to complete the SIC, unless special circumstances occur such as hospitalization or ventilation, in which case the reason for not completing the SIC should be recorded by site staff in the clinical database If signs and symptoms are still ongoing on **COVID-19 Day 3-5**, collection of SIC will be continued until AT LEAST 14 days after onset UNLESS both **COVID-19 Day 1-2** and **COVID-19 Day 3-5** are both negative. If either of the swabs is positive or the result is unknown AND the participant is beyond 14 days after onset of symptoms, the SIC can be stopped after 2 days without signs and symptoms.
   If a participant is unable to complete the eCOA, a study staff member or the participant's caregiver can collect information on the participant's behalf as detailed in Section 8.1.2.
   Participant should measure body temperature daily (oral route preferred, or in accordance with the local standard of care) and record the highest temperature in the last 24 hours.
t. If the participant does not have symptoms at that time, he/she will only need to complete the (suspected) COVID-19 surveillance (symptom check).
u. Includes measurement of supine (preferably) systolic and diastolic blood pressure, heart rate, and respiratory rate [after at least 5 minutes rest] and body temperature. It is recommended that vital signs are measured before collection of nasal swabs and blood draws.
v. In case of COVID-19 like symptoms, the participant will be asked to measure blood oxygen saturation and pulse rate at home 3 times a day (preferably in the morning, at lunch time, and in the evening). The results will be recorded by the participant in the eCOA.
w. Data collected as part of the MRU will be recorded in the eCRF.
x. The MA-COV form (Appendix 7) will be provided to the participant at the first study visit and should be completed by the medical care provider or the study site personnel during medical visits for COVID-19 or COVID-19 complications.

Upon closure of the COVID-19 episode and procedures, all participants will fall back to the default Schedule of Activities, until the end of the study/early withdrawal. If the participant experiences new signs or symptoms suggesting possible COVID-19 at a later point in time, the participant would re-start the COVID-19 procedures from COVID-19 Day 1 onwards. For participants experiencing COVID-19 episodes close to the end of their study participation: follow COVID episode procedures up to their EoT visit. For example, if an episode starts -4 days prior to scheduled EoT visit, complete **COVID-19 Day 1- 2** and **COVID-19 Day 3-5** procedures as much as possible, and then stop study procedures.

COVID-19 = coronavirus disease-2019; eCOA = electronic clinical outcome assessment; eCRF = electronic case report form; EoT= end of trial, ePRO = electronic patient-reported outcome; MA-COV = medically-attended COVID-19; MRU = medical resource utilization; SARS-CoV-2 = severe acute respiratory syndrome coronavirus-2; SIC = Symptoms of Infection with Coronavirus-19.

2. INTRODUCTION

In this study, immune responses against the original SARS-CoV-2 strain and leading variant of high consequence or concern[a] (and other variants of concern) induced by booster vaccination with Ad26.COV2.S ≥6 months after primary vaccination with Ad26.COV2.S or BNT162b2 will be compared to responses induced by Ad26.COV2.S or Pfizer BNT162b2 primary vaccination against the original SARS-CoV-2 strain and leading variant of high consequence or concern[b] (and other variants of concern).

Ad26.COV2.S Vaccine

Ad26.COV2.S (also known as VAC31518, JNJ-78436735) is a monovalent vaccine composed of a recombinant, replication-incompetent Ad26 vector, constructed to encode the S protein derived from a SARS-CoV-2 clinical isolate (Wuhan 2019, whole genome sequence NC_045512), stabilized in its prefusion conformation.

In response to the public health emergency caused by the global SARS-CoV-2 pandemic, the Ad26.COV2.S vaccine was authorized for emergency use in the United States (US) on 27 February 2021 and received a conditional Marketing Authorization approval in the European Union on 11 March 2021. On 12 March 2021, the Ad26.COV2.S vaccine was recommended for emergency use under the World Health Organization (WHO) Emergency Use Listing Procedure.

For the most comprehensive nonclinical and clinical information regarding Ad26.COV2.S refer to the latest version of the Investigator's Brochure (IB) (and Addenda, if applicable) for Ad26.COV2.S.

The term “study vaccine” throughout the protocol, refers to Ad26.COV2.S as defined in Section 6.1. The term "sponsor" used throughout this document refers to the entities listed in the Contact Information page(s), which will be provided as a separate document.

Study COV2008 is being conducted under the sponsorship of Janssen (Janssen Vaccines & Prevention B.V).

2.1. Study Rationale

The currently authorized Ad26.COV2.S vaccine directed against the original SARS-CoV-2 strain was associated with demonstrated clinical protective efficacy against COVID-19 (Sadoff 2021b). This study will assess the reactogenicity, safety, and immunogenicity of a booster dose of Ad26.COV2.S in adults ≥18 years of age, who have previously received primary vaccination with Ad26.COV2.S or the Pfizer mRNA-based vaccine BNT162b2.

[a] As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

The purpose of the study is to demonstrate that:

- The humoral neutralizing immune response elicited by booster vaccination with Ad26.COV2.S in participants who previously received primary vaccination with Ad26.COV2.S or BNT162B2 is non-inferior (NI) to the response elicited by primary vaccination with Ad26.COV2.S.

For Cohort 2 study arms, formal NI testing will be conducted by pooling 28 days post-dose 1 VNA data across groups 1, 2 and 3 (Cohort 1) compared to the 14 days post booster VNA data in the study arm of interest (groups 4, 5 or 6).

The Ad26.COV2.S booster will be assessed at dose levels of 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp.

Non-inferiority Testing

The neutralizing antibody response observed 14 days after booster vaccination will be the basis for NI evaluations. The primary purpose of the study is to assess whether individuals who received vaccination against the originally dominant strain with Ad26.COV2.S (Cohort 1), or with BNT162b2 (Cohort 2) can be adequately boosted by the standard dose level (5×10^{10} vp/dose), or lower dose levels (2.5×10^{10} vp and 1×10^{10} vp) of Ad26.COV2.S. Note: Analysis of Cohort 1 data can be completed prior to analysis of Cohort 2 data.

See Section 4.3 for further information of dose selection.

Other Evaluations

The study will assess the neutralization ability of the antibodies induced by the vaccine against the heterologous virus, as well the humoral immune response against other SARS-CoV-2 variants of concern. This includes, but is not limited to, the B.1.1.7 variant which first emerged in the United Kingdom [Leung 2021] and variants of the P.1 lineage which first emerged in Brazil [Faria 2021].

The study design and NI assessment are in line with guidance from the Food and Drug Administration (FDA 2021b) and the European Medicines Agency (EMA 2021) for the evaluation of a modified vaccine against a variant of SARS-CoV-2 in view of its authorization under Emergency Use Authorization or conditional Marketing Authorization.

2.2. Background

2.2.1. SARS-CoV-2 Virology and COVID-19 Disease Burden

SARS-CoV-2 is an enveloped, positive-sense, single-stranded ribonucleic acid (RNA) Betacoronavirus (Coronaviridae Study Group of the International Committee on Taxonomy of Viruses 2020; Wu 2020). In late December 2019, investigation of a cluster of pneumonia cases of unknown origin in Wuhan, China resulted in identification of a novel coronavirus (Chen N 2020; Li 2020) from the family Coronaviridae (Lu 2020; WHO 2020c). Phylogenetic analysis of the complete viral genome revealed that the virus, SARS-CoV-2, is part of the subgenus Sarbecovirus of the genus Betacoronavirus (Lu 2020; Zhou 2020). The virus is distinct from both severe acute

respiratory syndrome coronavirus (SARS-CoV) and Middle East respiratory syndrome coronavirus (MERS-CoV), although closely related.

SARS-CoV-2 is a highly transmissible and pathogenic coronavirus that has spread rapidly and globally since its emergence. The WHO declared that the outbreak constituted a public health emergency of international concern on 30 January 2020 and declared the outbreak to be a pandemic on 11 March 2020 (WHO 2020a; WHO 2020c). As of 17 April 2021, approximately 140,123,393 global cases of COVID-19 and approximately 3,002,736 COVID-19-related deaths have been reported worldwide. Within the United States alone, as of 17 April 2021, approximately 31,584,046 cases of COVID-19 and approximately 566,406 COVID-19-related deaths COVID-19 have been reported since the start of the outbreak (Johns Hopkins CSSE 2021). Within the European Union (EU)/European Economic Area (EEA) countries, as of 11 February 2021, a total of 20,478,718 cases of COVID-19 and 495,672 COVID-19-related deaths were reported (ECDC 2021).

During the COVID-19 pandemic, several new SARS-CoV-2 variants of concern emerged in the United Kingdom (B.1.1.7 lineage), in Brazil (P.1 lineage), in the Republic of South Africa (B.1.351 lineage), and in India (B.1.617 lineage) and new variants of interest (eg, B.1.427/B.1.429 lineage in California) continue to emerge, which may spread globally. The emergence of SARS CoV-2 variants with multiple mutations in the S protein have raised concerns because of their increased transmission rates, more severe disease (increased hospitalizations or deaths), and because of the possibility that current COVID-19 vaccines authorized under Emergency Use Authorization (EUA) or otherwise in clinical development will provide reduced protection against these variants (CDC 2021c; Rambaut 2020; Tegally 2020). For example, data suggest that the B.1.351 variant is not neutralized by some monoclonal antibodies directed to the SARS CoV-2 S protein and is resistant to neutralization by plasma from individuals previously infected with 'Wuhan-like' SARS-CoV-2 (Wibmer 2021). Although, data obtained to date suggest that the impact on neutralization by convalescent and post-vaccination sera is minimal to moderate (CDC 2021c).

A wide range of symptoms have been reported in association with SARS-CoV-2 infection. Symptoms usually appear 2 to 14 days (most commonly around 5 days) following exposure, with dyspnea and pneumonia typically developing within 8 days from onset of symptoms. Clinical manifestations of disease range from mild symptoms to severe illness or death usually associated with acute respiratory distress syndrome (ARDS) and respiratory failure. Multiple organ failure has also been reported in some COVID-19 cases (CDC 2020b; Guan 2020; Linton 2020; US San Diego Health 2020; WHO 2020b). Individuals aged 60 years or older as well as younger adults with serious comorbidities, such as cardiovascular disease, diabetes, hypertension and underlying pulmonary disease, are subject to the highest incidence of morbidity and mortality (CDC 2020h; Garg 2020; Verity 2020), with an increased risk of severe illness related to COVID-19 infection resulting in hospitalization, admission to the intensive care unit (ICU), intubation or mechanical ventilation, or death.

SARS-CoV-2 infection is often asymptomatic or associated with mild to moderate self-limiting symptoms such as fever, dry cough, myalgia, and fatigue (Huang C 2020; Chen G 2020). However, a subset of patients, especially younger adults, with severe SARS-CoV-2 infection develop a clinically severe hyperinflammatory state known as cytokine storm (CS) that is associated with elevated levels of the cytokines IL-6, and to a lesser extent IL-10 and TNF-α, as well as severe T cell lymphopenia and coagulopathy for which pulmonary involvement such as ARDS is a principal feature, resulting in a requirement for mechanical ventilation, and often death (Copaescu 2020). A multisystem inflammatory syndrome (MIS) that appears distinct from the adult CS and exhibiting features similar to atypical Kawasaki disease, has also been observed in a subgroup of previously healthy children following acute SARS-CoV-2 infection (CDC 2020g; Riphagen 2020).

Histopathological changes in patients with COVID-19 occur mainly in the lungs, with evidence of bilateral diffused alveolar damage. SARS-CoV-2 binds to the angiotensin-converting enzyme 2 (ACE2) receptor expressed on epithelial cells in the upper respiratory tract and alveolar epithelial cells in the lungs, eliciting a powerful cytokine mediated immune response that results in ARDS and respiratory failure, the main cause of death in patients with COVID-19 (Hu 2021). Lymphocytes expressing the ACE2 receptor may also be directly infected by the SARS-CoV-2 virus following sequestration at sites of infections such as the lung (Huang I 2020). Mild thrombocytopenia has been detected in 58-95% of severe cases of COVD-19, with severe thrombocytopenia only rarely reported. A meta-analysis of 7,613 COVID-19 patients revealed that patients with severe disease had lower platelet counts, often associated with increased mortality (Wool 2021).

To date, there are no proven effective therapies for COVID-19 or antivirals against SARS-CoV-2, although preliminary data suggests that some treatments may confer benefits to certain subpopulations of patients. A number of clinical studies to evaluate potential therapies for COVID-19 are currently ongoing. Vaccination remains the most effective method for a long-term strategy for prevention and control of COVID-19 in the foreseeable future. The SARS-CoV-2 virus that is responsible for COVID-19 is highly contagious and poses a severe threat to global health. SARS-CoV-2 has been shown to be highly pathogenic. The emergence of new variants with increased transmissibility that have the potential to cause more severe disease is a major concern.

2.2.2. Ad26.COV2.S

Nonclinical Pharmacology

Nonclinical pharmacology of Ad26.COV2.S was evaluated in murine, rabbit, Syrian hamster, and non-human primate (NHP) animal models for immunogenicity, including assessment of immunological parameters relevant to the theoretical risk of vaccine-associated enhanced respiratory disease (VAERD). In addition, vaccine efficacy of Ad26.COV2.S including lung histopathology assessment was evaluated in Syrian hamsters and NHPs. Details are provided in the IB (IB Edition 4 Ad26.COV2.S 2021 and its addenda).

Nonclinical Safety

Biodistribution

To assess distribution, persistence, and clearance of the Ad26 viral vector platform, intramuscular (IM) biodistribution studies have been conducted in rabbits using an Ad26-based human immunodeficiency virus (HIV) vaccine, Ad26.ENVA.01 (Ad26 vector encoding the Clade A envelope protein of HIV type 1), and an Ad26-based respiratory syncytial virus (RSV) vaccine, Ad26.RSV.preF (Ad26 vector encoding the prefusion conformation-stabilized F protein [pre-F] of RSV A2 strain). No pharmacokinetic or biodistribution studies have been conducted with the initial vaccine Ad26.COV2.S or the modified vaccine A26.COV2-S.02 specifically. The biodistribution studies showed a pattern of limited distribution of the Ad26 vector; Ad26 vector DNA was primarily detected at the site of injection, draining lymph nodes and (to a lesser extent) the spleen. Clearance (ie, reflected by a downward trend in number of positive tissues and vector copies over time, to levels close to, or below, the respective detection limits) of the Ad26 vector from the tissues was observed, indicating that the vector does not replicate and/or persist in the tissues. Both Ad26 vectors showed a comparable biodistribution profile despite carrying different antigen transgenes.

As biodistribution is considered dependent on the viral vector platform and not on the transgene insert, the biodistribution results obtained with Ad26.ENVA.01 and Ad26.RSV.preF are considered sufficient to inform on the biodistribution profile of Ad26.COV2.S when administered via the same (ie, IM) route.

Toxicology

In a GLP-compliant repeat-dose toxicity and local tolerance study in rabbits (TOX14382), Ad26.COV2.S was well tolerated when administered on 3 occasions over 4 weeks (ie, every 2 weeks) at 1×10^{11} virus particles (vp)/dose. The observed changes were related to a normal, anticipated (local and systemic) immunologic response to vaccination, and consisted clinically of (rare) transient local injection site dermal reactions, with transient minimal hyperthermia and minimal body weight loss or lower body weight gain after injection. This was associated with a transient (acute phase/immune) response in clinical pathology parameters, characterized by increases in plasma proteins and white blood cell counts. Microscopic pathology findings of minimal to slight inflammation and hemorrhage were observed at the injection sites, along with increased lymphoid cellularity of germinal centers in popliteal and iliac lymph nodes and the spleen, which is consistent with an immune response to the vaccine administration. Overall, the findings were considered non-adverse and were partially or completely reversible after a 3-week treatment-free period. Details are provided in the IB (IB Edition 4 Ad26.COV2.S 2021).

Reproductive and Developmental Toxicology

There was no harmful effect of Ad26.COV2.S with respect to female reproductive toxicity and fertility as assessed in a GLP combined EF-PPND toxicity study in the rabbit (TOX14389), in which Ad26.COV2.S was administered on 3 occasions (ie, 7 days prior to mating, followed by one vaccination during early gestation [GD6] and one vaccination during late gestation [GD20]) at $1\times10^{\mathbf{11}}$ vp/dose. No adverse effects were observed on reproductive performance, fertility, ovarian

and uterine examinations, or parturition. In addition, there was no adverse effect of vaccination on fetal body weights, external, visceral and skeletal evaluations, or on postnatal development of the offspring. The parental females as well as their fetuses and offspring exhibited SARS-CoV-2 S protein-specific antibody titers, indicating that maternal antibodies were transferred to the fetuses during gestation. Further details are provided in the IB (IB Edition 4 Ad26.COV2.S 2021).

Clinical Studies

At the time of protocol writing, Phase 1/2a (COV1001, COV1002) and Phase 2a (COV2001) clinical studies to assess the safety, reactogenicity, and immunogenicity of Ad26.COV2.S, as well as Phase 3 (COV3001, COV3009) clinical studies to assess the efficacy of Ad26.COV2.S in the prevention of molecularly confirmed moderate to severe/critical COVID-19 in adults are ongoing and data from interim analyses from these studies, except COV3009, are available.

Efficacy

A single dose of Ad26.COV2.S at 5×10^{10} vp protects against COVID-19 in adults ≥18 years of age, including adults ≥60 years of age. Based on the primary efficacy analysis of the pivotal Phase 3 study COV3001, including 19,630 participants who received Ad26.COV2.S and 19,691 participants who received placebo, vaccine efficacy (adjusted 95% confidence interval [CI]) for the co-primary endpoints against molecularly confirmed moderate to severe/critical COVID-19 in participants who were seronegative at time of vaccination was 66.9% (59.03; 73.40) when considering cases from at least 14 days after vaccination and 66.1% (55.01; 74.80) when considering cases from at least 28 days after vaccination. Ad26.COV2.S has been shown to be effective against all symptomatic COVID-19, highly effective in the prevention of severe/critical COVID-19 (particularly in prevention of hospitalization and death, across all countries and all ages), and effective against newly emerging strains, such as the 20H/501Y.V2 strain.

Safety

The most extensive safety information of the single-dose regimen of Ad26.COV2.S at 5×10^{10} vp is available from ≥43,000 participants, 18 years of age and above, including adults 60 years of age and above, enrolled in the ongoing pivotal Phase 3 study COV3001. A 2-dose vaccine regimen of Ad26.COV2.S at 5×10^{10} vp is currently under evaluation in the ongoing Phase 3 study COV3009. Supportive safety information (adverse events [AEs], clinical laboratory abnormalities, vital signs, and physical examination findings) is available from the interim analysis of the Phase 1 and 2 studies COV1001, COV1002, and COV2001. In COV2001, Ad26.COV2.S is being evaluated at a range of doses and intervals (1.25×10^{10} vp, 2.5×10^{10} vp, 5×10^{10} vp, and 1×10^{11} vp) in adolescents 12 to 17 years of age, adults 18 to 55 years of age and ≥65 years of age.

Ad26.COV2.S has an acceptable safety and reactogenicity profile when administered as a single-dose or as a 2-dose regimen in adults ≥18 years of age, including adults ≥60 years of age. No significant safety issues were identified. In general, a lower reactogenicity profile was observed in older adults compared to younger adults.

Overall, up to 22 January 2021, no safety concerns were identified after vaccination with Ad26.COV2.S as a single-dose or 2-dose vaccine regimen at dose levels up to 1×10^{11} vp. There

was a trend towards a decrease in the frequency of solicited local and systemic AEs (reactogenicity) with a decreasing dose level of Ad26.COV2.S (from 1×10^{11} vp to 2.5×10^{10} vp). Reactogenicity was demonstrated to be transient and most solicited AEs, including pyrexia, generally resolved in 1 to 2 days post-vaccination. In the ongoing Phase 3 study COV3001, out of over 43,000 participants, 19 deaths were reported, 3 in the Ad26.COV2.S group and 16 in the placebo group, all of which were considered unrelated to the study vaccine by the investigator. No AEs with a fatal outcome have been reported in the supportive Phase 1 and Phase 2 studies COV1001, COV1002 and COV2001. In the COV3001 study, at the time of the primary analysis, 0.4% participants in the Ad26.COV2.S group and 0.6% participants in the placebo group reported 1 or more serious adverse events (SAEs). Ten (<0.1%) participants reported SAEs that were considered to be related to the study vaccine: 7 SAEs were reported in 7 participants in the Ad26.COV2.S group: Grade 4: Guillain-Barré syndrome and pericarditis (one of each); Grade 3: radiculitis brachial, post-vaccination syndrome, and Type IV hypersensitivity (one of each); Grade 2: facial paralysis (Bell's Palsy - 2 cases reported); 3 SAEs were reported in 2 participants in the placebo group: Grade 4: deep vein thrombosis (DVT) (one participant); Grade 3: Epstein-Barr virus infection and atrial flutter (both were reported in the same participant). No participants in either vaccine group (Ad26.COV2.S or placebo) were withdrawn from the study due to AEs. Across studies COV1001, and COV1002, COV2001, 2 participants reported SAEs which were considered to be related to the study vaccine (both occurred in study COV1001): Grade 3 pyrexia and Grade 2 multiple sclerosis. Early discontinuations of vaccination or the study due to (S)AEs were infrequent in all groups.

Since then, 6 cases of severe allergic reactions were reported in study COV3012 (Sisonke [Together]), an open-label, single-arm Phase 3b vaccine implementation study sponsored by the South African Medical Research Council and conducted in collaboration with Janssen, which is currently ongoing in the Republic of South Africa. One case met the Brighton Collaboration case definition of anaphylaxis with level 2 of diagnosis certainty (Rüggeberg 2007). The review of the cases of severe allergic reactions reported from this study identified a plausible causal relationship between the administration of the Ad26.COV2.S vaccine and the occurrence of severe allergic reactions including anaphylaxis.

Cases of thrombotic events with thrombocytopenia have very rarely been observed in individuals who received Ad26.COV2.S. At the time of primary analysis of study COV3001 (Data lock point 22 January 2021), one case of cerebral venous sinus thrombosis (CVST) with thrombocytopenia had been reported which triggered a temporary pause in vaccinations across Ad26.COV2.S clinical studies. One additional case of DVT with thrombocytopenia was since reported in study COV3001. As of 17 April 2021, 6 spontaneous reports of CVST with thrombocytopenia and 1 spontaneous report of DVT with thrombocytopenia have been reported from the US in the context of the routine vaccination programs, one of which had a fatal outcome. At that time, more than 7.9 million people had received Ad26.COV2.S in the US.

Immunogenicity

Across studies COV1001, COV1002 and COV2001, a single dose of Ad26.COV2.S was shown to elicit SARS-CoV-2 neutralizing antibodies in both the 18 to 55 years age group and in adults

≥65 years of age. In studies COV1001 and COV3001, SARS-CoV-2 S protein binding antibodies were elicited in the vast majority of 18 to 55 years age group and in adults ≥65 years of age.

In study COV1001, Ad26.COV2.S induced CD4+ T cell responses with a Th1 dominant phenotype and CD8+ T cell responses in 18-55 and above 65 years of age participants.

Refer to the latest IB for more details on the ongoing clinical studies with Ad26.COV2.S.

2.2.3. Ad26-based Vaccines

Safety of Ad26-vectored vaccines has been evaluated in adults in clinical studies by the sponsor. Replication-incompetent Ad26 is being used as a vector in the development of vaccines against diseases such as malaria, RSV, HIV, Zika virus, filovirus, and HPV, and has been used in the now licensed Ebola virus vaccine (Zabdeno/Ad26.ZEBOV) and for emergency use licensed COVID-19 vaccine (Ad26.COV2.S).

As of 17 April 2021, Ad26-based vaccines developed by the sponsor have been administered to more than 8 million participants. The majority of these participants are enrolled in an ongoing immunization campaign in the United States (COVID-19 Vaccine Program Campaign) (more than 7.9 million) and an ongoing Ebola vaccine study in the Democratic Republic of the Congo and an ongoing immunization campaign in Rwanda (UMURINZI Ebola Vaccine Program campaign) (more than 156,000 to be updated with cut-off 17 April).

Overall, the Ad26-based vaccines were well tolerated irrespective of the antigen transgene, without significant safety issues. Recently, rare disorders including blood clots in combination with low platelets have been observed after vaccination with Ad26-based COVID-19 vaccines, including the Ad26.COV2.S vaccine (refer to Section 2.3.1 for more details).

2.2.4. mRNA-based SARS-CoV-2 Vaccines

The Pfizer-BioNTech COVID-19 vaccine, BNT162b2, is a lipid nanoparticle-formulated, nucleoside-modified RNA vaccine encoding a prefusion stabilized, membrane-anchored SARS-CoV-2 full-length spike protein (Polack 2020).

The vaccine was evaluated in study NCT04368728 in which a total of 21,720 participants received injections with BNT162b2 and 21,728 with placebo. A two-dose regimen of BNT162b2 conferred 95% protection against COVID-19 in participants aged ≥16 years. Safety over a median of 2 months was similar to other viral vaccines, with short-term, mild-to-moderate injection site pain, fatigue, and headache. The incidence of SAEs was low and was similar across the vaccine and placebo groups (Polack 2020).

The BNT162b2 vaccine is currently available in the U.S. under an Emergency Use Authorization (EUA) granted by the FDA on December 11, 2020.

2.3. Benefit-Risk Assessment

More detailed information about the known and expected benefits and risks of Ad26.COV2.S may be found in the most recent version of the IB and addenda.

2.3.1. Risks Related to Study Participation

The following potential risks for Ad26.COV2.S will be monitored during the study and are specified in the protocol:

Risks Related to Ad26.COV2.S

At the time of initial protocol writing, efficacy, immunogenicity and safety data were available from the ongoing clinical studies COV1001, COV1002, COV2001, COV3001, and COV3009 (see Section 2.2 for details).

Sites should advise participants that side effects include fever as well as injection site pain, headache, fatigue, myalgia, and nausea per the current ICF; however, the occurrence of fever appears to be more common in younger adults and can be severe.

Based on recent data (see Section 2.2.2 for details), anaphylaxis is considered an important identified risk for Ad26.COV2.S. Individuals should be observed by a healthcare provider after vaccination per protocol requirements.

Thrombosis in combination with thrombocytopenia (thrombosis with thrombocytopenia syndrome [TTS]), in some cases accompanied by bleeding, has been observed very rarely following vaccination with Ad26.COV2.S (see Section 2.2.2 for details). Reports include severe cases of venous thrombosis at unusual sites such as CVST, splanchnic vein thrombosis, and arterial thrombosis, in combination with thrombocytopenia. These cases occurred approximately 1-2 weeks following vaccination, mostly in women under 60 years of age. Thrombosis in combination with thrombocytopenia can be fatal. The exact physiology of TTS is unclear. TTS is considered an important identified risk for Ad26.COV2.S. Participants should be instructed to seek immediate medical attention if they develop symptoms such as shortness of breath, chest pain, leg swelling, persistent abdominal pain, severe or persistent headaches, blurred vision, skin bruising and/or petechiae beyond the site of vaccination. The medical management of thrombosis with thrombocytopenia is different from the management of isolated thromboembolic diseases. Study site personnel and/or treating physicians should follow available guidelines for treatment of thrombotic thrombocytopenia (eg, from the American Society of Hematology 2021; British Society of Haematology - Expert Haematology Panel 2021, and the CDC 2021d). The use of heparin may be harmful and alternative treatments may be needed. Consultation with a hematologist is strongly recommended. Management of the participant should not be delayed by decision-making of the AESI Adjudication Committee. Due to the possibility of the occurrence of TTS after vaccination with Ad26.COV2.S, additional reporting and data collection procedures have been included in the study for thrombotic events, thrombocytopenia, and TTS (see Section 8.4.7), which may facilitate diagnosis and clinical management of the event.

Guillain-Barré syndrome (GBS) has been reported very rarely following vaccination with Ad26.COV2.S. As a result, it is considered an identified risk. Investigators should be alert to GBS signs and symptoms to facilitate diagnosis, and to initiate adequate supportive care and treatment.

For the most comprehensive nonclinical and clinical information regarding Ad26.COV2.S, refer to the most recent version of the IB and addenda.

Risks Related to Ad26.COV2.S Administration after Primary Vaccination with mRNA Vaccines

To date, no clinical data are available for Ad26.COV2.S vaccination after primary vaccination with BNT162b2.

Risks Related to Adenoviral-vectored Vaccines

The clinical AdVac® safety database (report version 6.0, dated 20 April 2021, cut-off date 31 December 2019) contains pooled safety data from 32 Janssen-sponsored clinical studies with Ad26 vaccine candidates: Ad26.ZEBOV (Ebola; 10 studies), Ad26.ENVA.01, Ad26.Mos.HIV and Ad26.Mos4.HIV (HIV; 8 studies), Ad26.CS.01 (malaria; 1 study), Ad26.RSV.FA2 and Ad26.RSV.preF (RSV; 10 studies), and Ad26.Filo (filovirus; 1 study), Ad26.ZIKV.001 (Zikavirus; 1 study), and Ad26.HPV16 and Ad26.HPV18 (human papillomavirus [HPV]; 1 study). In these studies, 8,152 adult participants received at least 1 vaccination with an Ad26-based vaccine. The AdVac® safety database report includes data only from studies for which the database had been locked for either the interim, primary, or final analysis for which the sponsor has been fully unblinded at the cut-off date of this report. Overall, the Ad26-based vaccines were well tolerated, without significant safety issues identified.

The majority of solicited local and systemic AEs were of mild or moderate severity and usually started within 1 to 2 days after vaccination. Most of the events resolved within 1 to 3 days.

In adults, the most frequently reported solicited local AE was injection site pain (57.0% of Ad26 participants, compared with 19.2% of placebo participants). All other solicited local AEs were experienced by less than 25% of adult participants. Severe injection site pain was experienced by 1.0% of adult Ad26 participants and 0.1% of adult placebo recipients.

There was a trend toward an increase in the frequency of some local AEs with an increase in Ad26 dose, ie, injection site pain (18.7% of participants at the 0.8×10^{10} vp dose level, 38.7% of participants at the 2×10^{10} vp dose level, 52.0% of participants at the 5×10^{10} vp dose level, and 77.1% of participants at the 1×10^{11} vp dose level), and to a lesser extent injection site swelling (6.7%, 2.7%, 9.3%, and 17.6%, respectively). Injection site warmth was not collected at the 0.8×10^{10} vp and the 2×10^{10} vp dose level. The frequency of injection site warmth at the 5×10^{10} vp and the 1×10^{11} vp dose level was 19.5% and 26.7%, respectively. This trend needs to be interpreted with caution since the participants in the lower dose groups (0.8×10^{10} vp and 2×10^{10} vp dose level) were all from a single study (VAC52150EBL3002), and the majority of the participants in the highest dose group (1×10^{11} vp dose level) were also from a single study (VAC18193RSV2003).

The most frequently reported solicited systemic AEs (ie, reported in more than 30% of participants) for adult Ad26 participants were malaise (53.8%), fatigue (47.1%), headache (42.6%), and myalgia (37.6%), all of which were more frequent for Ad26 participants compared with placebo (36.4%, 25.8%, 23.6%, and 14.6% of placebo participants, respectively). Most of these events were considered related to the study vaccine. Pyrexia (9.2%) and vaccine-related pyrexia (8.4%) were also reported more frequently after administration of an Ad26-based vaccine compared with placebo (2.5% and 2.0%, respectively).

The majority of solicited systemic AEs were of mild or moderate severity. For adults, 6.3% of Ad26 participants and 1.6% of placebo participants reported severe solicited systemic AEs, mostly malaise, fatigue, chills, headache, and myalgia. Other severe solicited systemic AEs were reported in less than 1.5% of adult Ad26 participants.

The most frequently reported unsolicited AE in adult Ad26 participants was upper respiratory tract infection (4.5% vs. 5.2% in adult placebo participants). The most frequently reported unsolicited AEs considered related to the vaccine were neutropenia (0.7% of adult Ad26 participants vs. 0.3% of adult placebo participants) and dizziness (0.6% vs. 0.1%, respectively).

General Risks Related to Vaccination

In general, IM injection may cause local itching, warmth, pain, tenderness, erythema/redness, induration, swelling, arm discomfort, or bruising of the skin. Participants may exhibit general signs and symptoms associated with IM injection of a vaccine and/or placebo, including fever, chills, rash, myalgia, nausea/vomiting, headache, dizziness, arthralgia, general itching, and fatigue. These side effects will be monitored, but are generally short-term. Instructions regarding use of antipyretic medication can be found in Section 6.7.

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 (see above risks related to Ad26.COV2.S). Severe reactions are rare. Participants with a known allergy, or history of anaphylaxis or other serious adverse reactions to vaccines or their excipients (including specifically the excipients of the study vaccine) will be excluded from the study.

After vaccination, participants will remain at the study site for at least 30 minutes and will be closely observed by study staff. Necessary emergency equipment and medications must be available in the clinic to treat severe allergic reactions.

Pregnancy and Birth Control

The effect of the study vaccine on a fetus or on a nursing baby is unknown.

Given the limited number of incident pregnancies in the clinical studies with Ad26-based vaccines in the AdVac® safety database report (HIV vaccine: 20 pregnancies in participants and 10 in partners of participants; Ebola vaccine: 32 pregnancies in participants and 13 in partners of participants), it is not possible at present to draw firm conclusions on the safety of the vaccines when administered around the time of conception or prior to the initiation of the pregnancies. There is currently no concerning pattern of AEs in the pregnancies initiated around the time of vaccination or after exposure to the Ad26-based vaccines in the Janssen vaccines clinical development programs.

Participants who are pregnant and breastfeeding are allowed to participate in the study.

Risks from Blood Draws

Blood draws may cause pain, tenderness, bruising, bleeding, dizziness, vasovagal response, syncope, and rarely, infection at the site where the blood is taken.

Theoretical Risk of Vaccine-associated Enhanced Respiratory Disease

Vaccine-associated enhanced disease (VAED) has been linked to a Th2-polarized immune response and inadequate induction of neutralizing antibody responses and it was characterized by enhanced lung histopathology in nonclinical challenge models of other coronaviruses or RSV (Agrawal 2016; Bolles 2011; Deming 2006; Honda-okubo 2015; Houser 2017). There is, therefore, a theoretical risk of VAED) including vaccine-associated enhanced respiratory disease (VAERD), for SARS-CoV-2 vaccines. The potential of Ad26.COV2.S to predispose for VAERD and VAED has been evaluated in nonclinical challenge models established by the sponsor and partners. Ad26.COV2.S immunized Syrian hamsters and NHP showed absence of enhanced lung pathology and clinical signs of disease compared with controls after SARS-CoV-2 inoculation, even under conditions of suboptimal immunity allowing breakthrough infection. Together with induction of neutralizing antibodies and a Th1 dominant immune response after Ad26.COV2.S dosing these data suggest that the theoretical risk of VAERD and VAED for Ad26.COV2.S is low (He 2021; van der Lubbe 2021). In both species, no signs of enhanced respiratory tract pathology were observed in animals dosed with Ad26.COV2.S compared with challenge control groups.

VAERD has so far not been reported in any of the clinical studies, including interim data reported from Phase 2/3 clinical trials of mRNA-based vaccines and an adenovector-based vaccine, expressing SARS-CoV-2 S protein as antigen (Baden 2020b; Polack 2020; Voysey 2021). Similarly, no cases of VAERD have been reported in any of the ongoing clinical studies conducted to date by the sponsor, including the Phase 1, Phase 2a and Phase 3 studies. The Ad26 vaccine vector platform has been assessed in several nonclinical and clinical studies so far, and uniformly induced potent humoral and cellular immune responses with a clear Th1 bias (Barouch 2018; Salisch 2019; van der Fits 2020; Widjojoatmodjo 2015). The Ad26.COV2.S vaccine candidate, which encodes the SARS-CoV-2 S protein, has been shown to induce S-protein binding antibodies and virus neutralizing antibodies in mice, rabbits, hamsters and NHP. Ad26.COV2.S was shown to induce a Th1 skewed immune response in mice and in NHP including aged NHP. Immunogenicity data from Study COV1001 demonstrated that a single vaccination with Ad26.COV2.S induces robust neutralizing and binding antibody, CD4+ T cell responses with a

Th1-dominant phenotype, and CD8+ T cell responses (Anywaine 2019; Barouch 2013; Barouch 2018; Sadoff 2021a).

Participants in the present study will be informed of the theoretical risk of disease enhancement in the informed consent form (ICF).

Unknown Risks

There may be other risks that are not known. If any significant new risks are identified, the investigators and participants will be informed.

2.3.2. Benefits of Study Participation

Participants may benefit from clinical testing and physical examination.

The efficacy, immunogenicity and safety data to date support a favorable benefit-risk profile for Ad26.COV2.S in the proposed indication, ie, active immunization to prevent COVID-19 caused by SARS-CoV-2 in adults ≥18 years of age (see Section 2.2). The overall evaluation of the benefit and risk balance for individual participants receiving Ad26.COV2.S in clinical studies and vaccination campaigns is ongoing.

2.3.3. Benefit-Risk Assessment of Study Participation

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:

- A single dose of Ad26.COV2.S protects against moderate to severe/critical COVID-19 as early as 14 days after vaccination as demonstrated in adults ≥18 years of age, including adults ≥60 years of age.

  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 the Schedule of Activities.

  After vaccination, participants will remain at the study site for at least 30 minutes 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 e-Diary to document solicited signs and symptoms. Details are provided in Section 8.3.

  The investigator or the designee will document unsolicited AEs and SAEs as indicated in Section 8.3, Section 8.4, and Appendix 4.

  TTS is considered to be an adverse event of special interest (AESI) (Section 8.4.7.1). Suspected AESIs (thrombotic events and thrombocytopenia [defined as platelet count below 150,000/µL (Brighton Collaboration 2021)]) must be reported to the sponsor

within 24 hours of awareness. Suspected AESIs will be followed up as described in the Schedule of Activities in Section 1.3.2.

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 clinically stable.

- An Independent Data Monitoring Committee (IDMC) for this compound has been assembled by the sponsor. The safety of this vaccine has been, and is being, reviewed by the IDMC under several study protocols. Post primary analysis, safety data from the study does not require routine presentation to the IDMC. Nevertheless, the IDMC will review safety data from this study on an ad hoc basis should any safety issues or concerns arise. The IDMC's responsibilities, authorities, and procedures are documented in the IDMC Charter.Several safety measures are included in this protocol to minimize the potential risk to participants, including the following:

  In each group in Cohort 2, 2 sentinel participants will be evaluated for safety before extending enrollment. The sentinel participants will be vaccinated at least 1 hour apart. Sentinel participants will come in for a study visit on Day 2 (approximately 24 hours post-vaccination) to collect safety data. The blinded 24-hour post-vaccination safety data in these sentinel participants will be reviewed by the principal investigator (PI) and sponsor's study responsible physician (SRP)/study responsible scientist (SRS). Randomization and vaccination of additional Cohort 2 participants will be halted until this 24-hour sentinel safety evaluation is completed. Refer to Section 4.1 for further details.

  Clinical laboratory assessments (see Appendix 2) will be performed as indicated in the Schedule of Activities.

  There are pre-specified pausing rules for all participants (see Section 6.8), that if met would result in pausing of further vaccinations, preventing exposure of new participants to study vaccine until the IDMC reviews all safety data.

  Reasons for Participant Discontinuation/Withdrawal are included in Section 7.

  Contraindications to vaccination are included in Section 5.5.

3. OBJECTIVES AND ENDPOINTS

Objectives	Endpoints
Primary	
Primary Objective 1a: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original

Objectives	Endpoints
induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level. ***If Primary Objective 1a is met, Primary Objective 1b will be tested.***	strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 100*(1-2*α)% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 100*(1-2*α)% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 1b: To demonstrate the NI of the neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, if feasible. ***If Primary Objective 1b is met, Primary Objectives 1c and 2a will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the leading variant of high consequence or concern*, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the leading variant of high consequence or concern*, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 100*(1-2*α)% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 100*(1-2*α)% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.

Objectives	Endpoints
Primary Objective 1c: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level. ***If Primary Objective 1c is met, Primary Objective 1d will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 1d: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, administered ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (1×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level). • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in responder rate (Ad26.COV2.S booster - Ad26.COV2.S primary) needs to be >-10%. AND

Objectives	Endpoints
	• The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT Ad26.COV2.S primary) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.
Primary Objective 2a: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2 ***If Primary Objective 2a is met, Primary Objective 2b will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain in serum samples of approximately 300 individuals, collected 2 weeks to 2 months after completing 2-dose primary vaccination with Pfizer BNT162b2 (further referred to as Pfizer BNT162b2 external samples). *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster - Pfizer BNT162b2 primary regimen [external samples]) needs to be > -10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/ post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2b: To demonstrate the NI of neutralizing antibody response to the leading variant of high consequence or concern* 14 days after booster vaccination	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) against the leading variant of high consequence or

Objectives	Endpoints
with Ad26.COV2.S at the 5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by 2-dose primary vaccination with Pfizer BNT162b2, if feasible ***If Primary Objective 2b is met, Primary Objectives 2c will be tested.***	concern* 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) against the leading variant of high consequence or concern* in Pfizer BNT162b2 external samples *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster - Pfizer BNT162b2 primary regimen [external samples]) needs to be > -10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2c: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2 ***If Primary Objective 2c is met, Primary Objective 2d will be tested.***	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2. • Serological response to vaccination and antibody titers (VNA) against the original strain, in Pfizer BNT162b2 external samples. *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster- Pfizer BNT162b2

Objectives	Endpoints
	primary regimen [external samples]) needs to be > -10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.
Primary Objective 2d: To demonstrate the NI of the neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, administered ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (1×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2. • Serological response to vaccination and antibody titers (VNA) against the original strain, in Pfizer BNT162b2 external samples . *Success criteria for NI:* • The lower limit of the 97.5% CI on the difference in seropositivity rate (Ad26.COV2.S booster- Pfizer BNT162b2 primary regimen [external samples]) needs to be > -10%. AND • The lower limit of the 97.5% CI on the ratio of neutralizing antibody GMTs (GMT Ad26.COV2.S booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) needs to be >2/3. AND • An estimated GMR (GMT Day 15 post-booster/GMT post Pfizer BNT162b2 primary regimen [external samples]) of >0.8 is required to conclude NI.

Objectives	Endpoints
Secondary	
To assess the safety and reactogenicity of Ad26.COV2.S administered at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels administered as booster vaccinations in adults.	• Solicited local and systemic adverse events (AEs) for 7 days after booster vaccination. • Unsolicited AEs for 28 days after booster vaccination. • Serious adverse events (SAEs) and adverse events of special interest (AESIs) throughout the study (from booster vaccination until end of the study).
To assess the neutralizing and binding antibody response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, induced by booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, in adults who have previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain, leading variant of high consequence or concern* AND other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination. • Antibodies binding to SARS-CoV-2 relevant variants of concern or individual SARS-CoV-2 proteins (eg, S and/or receptor-binding domain [RBD] proteins from the SARS-CoV-2 variants of concern) by ELISA and/or MSD.
To assess the neutralizing and binding antibody response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, induced by booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, in adults who have previously completed primary vaccination with Pfizer BNT162b.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain, leading variant of high consequence or concern* AND other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination. • Antibodies binding to SARS-CoV-2 relevant variants of concern or individual SARS-CoV-2 proteins (eg, S and/or receptor-binding domain [RBD] proteins from the SARS-CoV-2 variants of concern) by ELISA and/or MSD.
To assess previous or concomitant infection with SARS-CoV-2 at baseline.	Antibodies binding to the SARS-CoV-2 nucleocapsid (N) protein at Day 1 (N-serology).
Exploratory	
To assess the neutralizing antibody responses against the original strain, leading variant of high consequence or concern* and other relevant variants of concern, following Ad26.COV2.S booster vaccination in participants that previously received Ad26.COV2.S or Pfizer BNT122b2 as primary vaccination, compared observationally to antibody responses 14 days post primary vaccination	• Serological response to vaccination and antibody titers measured by VNA against the original strain, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively.

Objectives	Endpoints
in participants from Study COV3009 (2-dose schedule of Ad26.COV2.S spaced by 56 days).	• Serological response to vaccination and antibody titers measured by VNA against, leading variant of high consequence or concern*, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively. • Serological response to vaccination and antibody titers measured by VNA against other relevant variants of concern, 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10}vp dose level), after completing one-dose or 2-dose primary vaccination with Ad26.COV2.S or Pfizer BNT162b2, respectively. • Serological response to vaccination and antibody titers measured by VNA against the original variant, 14 days after completing Ad26.COV2.S primary vaccination as a 2-dose schedule spaced by 56 days (5×10^{10}vp dose level), in a subset of participants from study COV3009.
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp vs those receiving 2.5×10^{10} vp, in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (2.5×10^{10} vp or 5×10^{10} vp dose level).
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp vs those receiving 1×10^{10} vp, in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (1×10^{10} vp or 5×10^{10} vp dose level).
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination (5×10^{10} vp) in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and after booster vaccination at

Objectives	Endpoints
against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Pfizer BNT162b.	the 5×10^{10} vp dose level in participants primed with Pfizer BNT162b.
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses against the original strain induced by booster vaccination with 2.5×10^{10} vp in adults who previously completed primary vaccination with Pfizer BNT162b.	• Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination at the 5×10^{10} vp dose level in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and booster vaccination at the 2.5×10^{10} vp dose level in participants primed with Pfizer BNT162b.
Observational comparison of neutralizing antibody responses against the original strain induced by booster vaccination with 5×10^{10} vp in adults who previously completed single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level vs neutralizing antibody responses against the original strain induced by booster vaccination with 1×10^{10} vp in adults who previously completed primary vaccination with Pfizer BNT162b.	Serological response to vaccination and antibody titers, as measured by VNA, against the original strain 14 days and 28 days after Ad26.COV2.S booster vaccination at the 5×10^{10} vp dose level in participants primed with the 1-dose Ad26.COV2.S vaccine at the 5×10^{10} vp dose level, and booster vaccination at 1×10^{10} vp dose level in participants primed with Pfizer BNT162b.
To determine if any of the primary objectives that meet a non-inferiority criterion also meet a superiority criterion.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp OR 2.5×10^{10} vp OR 1×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S OR 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination.
To make observational comparisons between the immunologic response to booster vaccination and the primary vaccination regimen, in participants primed with the Pfizer BNT162b regimen (if	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the original strain, 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp OR 2.5×10^{10} vp OR 1×10^{10} vp dose level) after

Objectives	Endpoints
appropriate serum samples are available for testing).	completing 2-dose primary vaccination with Pfizer BNT162b2 • Serological response to vaccination and antibody titers (VNA) against the original strain, 28 days after completing 2-dose primary vaccination with Pfizer BNT162b2
To evaluate patient-reported outcomes (PROs) in relation to the presence of SARS-CoV-2 infection and the presence, severity and duration of COVID-19 signs and symptoms in participants who receive booster vaccination.	• Presence, severity and duration of COVID-19 signs and symptoms • Confirmation of SARS-CoV-2 infection by molecular testing • Evaluate all PCR and N ELISA confirmed cases of SARS-CoV-2 infections according to the charter of the Clinical Severity Adjudication Committee.
To explore the relative vaccine efficacy (rVE) of Ad26.COV2.S heterologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate, moderate to severe/critical, mild, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) compared to Ad26.COV2.S homologous booster vaccination at the same dose level (5×10^{10}vp, 2.5×10^{10}vp and 1×10^{10}vp).	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 nucleocapsid (N) protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate, moderate to severe/critically ill and severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.
To explore the rVE of different dose levels of Ad26.COV2.S homologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate to severe/critical, moderate, mild, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) as follows: 5×10^{10}vp versus 1×10^{10}vp dose level 2.5×10^{10}vp versus 1×10^{10}vp dose level	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate, moderate to severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.

Objectives	Endpoints
To explore the rVE of different dose levels of Ad26.COV2.S heterologous booster vaccination in the prevention of SARS-CoV-2 infection (severe/critical, moderate to severe/critical, moderate, mild, symptomatic, and asymptomatic infections, that are serologically and/or molecularly confirmed) as follows: 5×10^{10}vp versus 1×10^{10}vp dose level 2.5×10^{10}vp versus 1×10^{10}vp dose level	• Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein. • SARS-CoV-2 infections as detected by molecular testing (RT-PCR or equivalent). • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post vaccination in COV2008. • COVID-19 cases meeting the criteria for "moderate and moderate to severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.
If feasible, to investigate the effect of post booster cellular responses, mRNA profiles, neutralizing responses and/or other functional antibody responses on the probability of experiencing a COVID-19 event, moderate and moderate to severe/critically ill COVID-19 disease, or asymptomatic SARS-CoV-2 infection.	• Serological response to vaccination and antibody titers as measured by virus neutralization assay (VNA) titers against the reference strain and/or SARS-CoV-2 variants, by ELISA/MSD and/or other functional antibody assays, 14 days, 6 months and 1 year after homologous or heterologous booster vaccination with Ad26.COV2.S, if sufficient data are available. • Cellular response to vaccination as measured by flow cytometry, ELISPOT and/or transcriptomics, 14 days, 6 months and 1 year after homologous or heterologous booster vaccination with Ad26.COV2.S, if sufficient data are available. • COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition, with onset at least 14 days post booster vaccination in study COV2008, if sufficient data are available. • COVID-19 cases meeting the criteria for "moderate and moderate to severe/critically ill" COVID-19, with onset at least 14 days post vaccination in COV2008, if sufficient data are available. • Asymptomatic SARS-CoV-2 infection, with onset at least 14 days post vaccination in COV2008, if sufficient data are available.

Objectives	Endpoints
	• Analysis of gene expression by RNA transcript profiling and correlation with humoral and cellular immune responses.
In a subset, to assess the cellular immune response to the original strain, leading variant of high consequence or concern* and other relevant variants of concern, after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp dose levels, at baseline (Day 1, pre-booster vaccination) and 14 days, 6 months and 1 year after booster vaccination in adults. *This objective will be tested only if PBMC collection is feasible.*	T helper (Th) 1 and Th2 immune responses as assessed by: • Flow cytometry after SARS-CoV-2 S protein peptide stimulation of peripheral blood mononuclear cells (PBMC) and intracellular staining (ICS) including $CD4^+/CD8^+$, interferon gamma (IFNγ), interleukin (IL)2, tumor necrosis factor alpha (TNFα), IL-4, IL-5, IL13, and/or other Th1/Th2 markers. AND/OR • Dual or single IFNγ and IL-4 enzyme-linked immunospot (ELISpot) assay after stimulation of PBMCs with SARS-CoV-2 S protein peptides.
In a subset of participants, to assess the cellular immune response to the original strain, leading variant of high consequence or concern* and other relevant variants of interest, after booster vaccination with Ad26.COV2.S in adults who have completed a primary vaccination regimen with Ad26.COV2.S or BNT162b2, at baseline (Day 1, pre-booster vaccination) and 14 days, 6 months and 1 year after booster vaccination. *This objective will be tested only if PBMC collection is feasible.*	T helper (Th) 1 and Th2 immune responses as assessed by: • Flow cytometry after SARS-CoV-2 S protein peptide stimulation of peripheral blood mononuclear cells (PBMC) and intracellular staining (ICS) including $CD4^+/CD8^+$, interferon gamma (IFNγ), interleukin (IL)2, tumor necrosis factor alpha (TNFα), IL-4, IL-5, IL13, and/or other Th1/Th2 markers. AND/OR • Dual or single IFNγ and IL-4 enzyme-linked immunospot (ELISpot) assay after stimulation of PBMCs with SARS-CoV-2 S protein peptides.
To further explore the humoral immune response to Ad26.COV2.S booster vaccination in adults.	Exploratory analyses may include the following assays: • SARS-CoV-2 neutralization as assessed by alternative SARS-CoV-2 neutralization assays (VNA). • Adenovirus neutralization. • Functional and molecular antibody characterization including Fc-mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype.

Objectives	Endpoints
	• Epitope-specificity characterization of antibodies. • Cytokine profiling: Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma. • Passive transfer: Analysis of immune mediators correlating with protection against experimental SARS-CoV-2 challenge in a suitable animal model.
In a subset, to assess humoral and cellular responses (if feasible) 28 days following the second dose of Pfizer BNT162b2 against the original strain, leading variant of high consequence or concern* and other relevant variant strains for comparison to booster responses in the different regimens and the primary response to Ad26.COV2.S.	• Assay panels similar to those tested for the other groups will be utilized based on availability of serum and cells. Assessments performed with different assays to those utilized for other groups in this study may be utilized.
At one site (BIDMC), in-depth humoral and cellular immunogenicity assessment may be performed on participants enrolled at that site and a subset of participants enrolled at other sites.	Exploratory analyses may include the following assays: • SARS-CoV-2 neutralization as assessed by alternative SARS-CoV-2 neutralization assays (VNA). • Adenovirus neutralization. • Functional and molecular antibody characterization including Fc-mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype. • Epitope-specificity characterization of antibodies. • Cytokine profiling: Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma. • Passive transfer: Analysis of immune mediators correlating with protection against experimental SARS-CoV-2 challenge in a suitable animal model. • Analysis of gene expression by RNA transcript profiling

Objectives	Endpoints
To characterize and establish correlations between mRNA profiling and immunologic responses to the booster vaccinations.	• Analysis of gene expression by RNA transcript profiling and correlation with humoral and cellular immune responses.
To explore markers of inflammation and upregulation of pathways that may be involved in the thrombosis with thrombocytopenia syndrome (TTS) rarely occurring after vaccination with Ad26.COV2.S.	• Descriptive comparison of RNA sequence profiles from participants receiving Ad26.COV2.S vaccination to the RNA profiles of participants receiving other Ad26-based vaccines.
Explore the relationship between humoral and cellular (if feasible) responses in relationship to the time between primary and booster immunization	• In silico modelling of the humoral and cellular immune responses elicited by Ad26.COV2.S as primary regimen and by Ad26.COV2.S as booster, over time.
To assess the vaccine recipient's risk of developing thrombotic events after Ad26.COV2.S administration.	• Platelet count pre-vaccination at Day 1 (all participants) and Day 29 post-vaccination (for participants with a suspected AESI). • Based on the clinical evaluation of the suspected AESI (eg, whether thrombocytopenia is observed with a thrombotic event), a panel of coagulation-related tests may be conducted on the stored pre-vaccination sample (retrospective test), on Day 29 post vaccination, and on the samples obtained as part of the AESI investigation).

* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

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

HYPOTHESIS

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively by vaccine group. See Section 9.1 for details of immunogenicity hypothesis testing.

4. STUDY DESIGN

4.1. Overall Design

This randomized, double-blind, parallel, multicenter study will assess the reactogenicity, safety and immunogenicity of a booster dose of Ad26.COV2.S in adults ≥18 years of age, who have previously received primary vaccination with Ad26.COV2.S or BNT162b2.

The primary immunogenicity endpoints are designed to sequentially show non-inferiority of the responses after boosting with various doses of Ad26.COV2.S in volunteers either primed with Ad26.COV2.S at a 5×10^{10} vp dose level, or Pfizer BNT162b2, vs the neutralizing antibody

responses to the primary 1-dose regimen of Ad26.COV2.S which has shown efficacy in a prospective randomized Phase 3 efficacy trial. This study will therefore attempt to show which booster regimens will induce neutralizing antibody responses non-inferior to those seen with the Janssen 1-dose regimen of Ad26.COV2.S which has demonstrated efficacy.

In Cohort 1 a target of approximately 770 participants who have received Ad26.COV2.S in study COV3001 will initially be randomized in a 1:1:1 ratio into 3 groups to receive a 1-dose booster vaccination regimen with Ad26.COV2.S until the group 3 is fully enrolled. Thereafter, randomization will continue in a 1:1 ratio in groups 1 and 2 as shown in Table 1. When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort 1 Group 1 participants. The other primary objectives will only be tested at the primary analysis.

In Cohort 2 a target of approximately 770 participants who have received primary vaccination with the Pfizer BNT162b2 vaccine will initially be randomized in a 1:1:1 ratio into 3 groups to receive a 1-dose booster vaccination regimen with Ad26.COV2.S until the group 6 is fully enrolled. Thereafter, randomization will continue in a 1:1 ratio in groups 4 and 5 as shown in Table 1.

Table 1: Schematic Overview of Study Design and Groups

Cohort	Group	N	Day 1 Vaccination
Cohort 1 (Ad26.COV2.S primary vaccination)	1	~ 330	Ad26.COV2.S at 5×10^{10} vp
	2	~ 330	Ad26.COV2.S at 2.5×10^{10} vp
	3	~ 110	Ad26.COV2.S at 1×10^{10} vp
Cohort 2 (BNT162b2 primary vaccination)	4	~ 330	Ad26.COV2.S at 5×10^{10} vp
	5	~ 330	Ad26.COV2.S at 2.5×10^{10} vp
	6	~ 110	Ad26.COV2.S at 1×10^{10} vp

In Cohort 2, the study vaccine will first be administered to 6 sentinel participants (2 participants per group), enrolled at the same study site, to monitor for unexpected severe adverse reactions. The sentinel participants will be vaccinated at least 1 hour apart and closely observed for a minimum of 30 minutes post-vaccination for the development of acute reactions. Each sentinel participant is scheduled to come to the site for a study visit on Day 2 (approximately 24 hours post-vaccination) to collect safety data, which will include solicited AEs (collected through e-Diary) and unsolicited AEs, AESIs, and SAEs. The collected data will be reviewed in a blinded manner by the PI and the sponsor's SRP/SRS. Randomization and vaccination of additional participants will be halted until the review of sentinel data is completed. In the absence of clinically significant findings from the review of 24-hour safety data from the sentinel participants, randomization and vaccination will continue.

Active surveillance for COVID-19-like signs and symptoms will occur, including a COVID-19 surveillance (symptom check) through the eCOA. All participants with COVID-19-like signs or symptoms meeting the prespecified criteria for suspected COVID-19 (see Section 8.1.1) and all participants with at least 1 positive RT-PCR test for SARS-CoV-2 within 5 days of symptom onset, should undertake pre-specified COVID-19 procedures (see Section 8.1.2).

Study Duration

The study duration from screening until the last follow-up visit will be approximately 1 year per participant. The study will consist of a 14-day screening phase, booster vaccination on Day 1, and follow-up visits up to 1 year after booster vaccination (Target Visit Day 361±21 days).

If a participant is unable to complete the study, but has not withdrawn consent, an early exit visit will be conducted. The end of study is considered as the last visit for the last participant in the study.

Study Procedures

For each group, safety will be assessed by collection of solicited local (at injection site) and systemic AEs, unsolicited AEs, AESIs, and SAEs. Other safety assessments include clinical laboratory assessments, vital signs measurements (pulse/heart rate, preferably supine systolic and diastolic blood pressure, respiratory rate, and body temperature) and physical examinations at the time points indicated in Section 1.3.

After booster vaccination, participants will remain under observation at the study site for at least 30 minutes for presence of any acute reactions and solicited events. Any solicited local or systemic AEs, unsolicited AEs, AESIs, SAEs, concomitant medications, and vital signs will be documented by study site personnel following this observation period. In addition, participants will record solicited signs and symptoms in an e-Diary for 7 days post-vaccination.

The occurrence of asymptomatic SARS-CoV-2 infection will be assessed by a non-S-protein assay (eg, nucleocapsid [N] protein ELISA and/or SARS-CoV-2 immunoglobulin assay that is dependent on the SARS-CoV-2 N protein) (see Section 8.1.1).

The reporting periods of AEs, AESIs, SAEs, and special reporting situations are detailed in Section 8.4. Reporting periods for concomitant therapy are outlined in Section 6.7.

A final safety and immunogenicity follow-up visit is foreseen 1 year after vaccination for all participants.

The first ~330 randomized participants in each of the cohorts will be assigned to 1 of 4 blood collection subsets (Subsets 1, 2, 3 and 4). Blood samples will be collected for immunogenicity assessments at selected timepoints as indicated in Table 2,

Table 3 and Section 1.3. Once the 4 blood collection subsets are enrolled, subsequent participants will not be assigned to a subset. Participants not assigned to a subset ('non-subset' in Table 2) will have immunogenicity samples taken at Day 1, Day 15, Day 120, Day 181 and Day 361.

Table 2: Blood Collection Schedule (Whole Blood) for Humoral and Cellular Immunogenicity Assessments

Cohort	Group	Day 1[ab]	Day 2[b]	Day 8[ab]	Day 15[ab]	Day 29[a]	Day 71	Day 120	Day 181	Day 361
1	1	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)	Non-Subset[c] (N=~380)	All	All
	2	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
	3	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
2	4	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)	Non-Subset[c] (N=~380)	All	All
	5	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All
	6	All	Subset 3 (N ~27)	Subset 1 (N ~27)	All	Subset 2 (N ~28)	Subset 3 (N ~27)		All	All

a. Selected timepoints include samples for PAXgene analysis.
b. Selected time points include samples for cytokine analysis (Subset 1, 2 and 3 only).
c. Day 120 blood draw for participants not allocated to a subset

Table 3: Blood Collection Schedule (PBMC[a]) for Cellular Immunogenicity Assessments

	Group	Day 1	Day 15	Day 181	Day 361
1	1	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	2	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	3	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
2	4	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	5	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)
	6	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)	Subset 4 (N ~28)

a. Blood collection for PBMC will be performed in a subset of participants in all groups (Subset 4), if PBMC analysis is feasible.

From external sources, the sponsor will obtain serum samples of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used to assess NI of the seropositivity rate induced by a Ad26.COV2.S booster following a primary Pfizer BNT162b2 regimen to the seropositivity post a primary Pfizer BNT162b2 regimen.

Further details about the immunogenicity assessments are provided in Section 8.1.5.

An IDMC has been commissioned for the Ad26.COV2.S program. Any significant safety information will be shared with the IDMC. After the primary analysis, data will be shared with the IDMC on an ad hoc basis, if there is a safety issue or concern. (see Section 6.8).

The planned primary and final analyses are detailed in Section 9.5. A diagram of the study design is provided in Section 1.2.

4.2. Scientific Rationale for Study Design

Dose Level Selection

The 5×10^{10} vp dose level was assessed for Ad26.COV2.S in study COV1001 and demonstrated robust immunogenicity together with acceptable safety and reactogenicity (see Section 4.3 for more details).

The study will assess 3 dose levels of Ad26.COV2.S booster vaccination: 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp .

Blinding, Control, Study Phase/Periods, Intervention Groups

Randomization will be used to minimize bias in the assignment of participants to vaccine groups, to increase the likelihood that known and unknown participant attributes (eg, demographic and baseline characteristics) are evenly balanced across vaccine groups, and to enhance the validity of statistical comparisons across vaccine groups. Blinded study vaccine will be used to reduce potential bias during data collection and evaluation of clinical endpoints.

Blinding will be guaranteed by the preparation of the study vaccine by an unblinded pharmacist or by other qualified study site personnel with primary responsibility for study vaccine preparation and dispensing, and by the administration of vaccine in a masked syringe by a blinded study vaccine administrator. Participants in each Cohort will be randomly assigned to 1 of 3 groups, based on a computer-generated randomization schedule prepared before the study by, or under the supervision of, the sponsor.

Biomarker Collection

For participants with a positive test result for SARS-CoV-2 infection, biomarker analysis (PAXgene, RNA-seq) will be performed for evaluation of COVID-19 cases and to explore potentially informative biomarkers, eg, those associated with severe COVID-19.

4.2.1. 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 that this study will be performed in adult participants who will receive no benefit from participation in the study, except for receiving vaccination against SARS-CoV-2 and compensation for their time and for the inconveniences that may arise from participation in the study. See Section 2.3 for details on potential and known benefits and risks, and for the safety measures taken to minimize risk to participants.

The total blood volume to be collected is considered to be an acceptable amount of blood to be collected over this time period from the population in this study based upon the US Department of Health and Human Services Office for Human Research Protections, and US FDA guidelines of 550 mL in any 8-week period (US FDA 1998; US DHHS 1998), and as well as the European Commission guidelines of 500 mL per donation and 3 L per consecutive 12 month period (EC 1998).

4.3. Justification for Dose

The 5×10^{10} vp dose level of Ad26.COV2.S was evaluated in study COV1001 (see also Section 2). However, recent data from study COV2001 suggest that a lower dose may be sufficient as a late booster. A lower booster dose has potential advantages, including a lower inflammatory immune response and lower incidence of resultant AEs. Therefore, 2.5×10^{10} vp and 1×10^{10} vp dose levels will also be assessed. Evaluation of the 2.5×10^{10} vp and 1×10^{10} vp dose levels will generate end-of-shelf-life data for the 5×10^{10} vp and 2.5×10^{10} vp dose levels, respectively.

4.4. End of Study Definition

End of Study Definition

The end of study is considered as the last visit for the last participant in the study. 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.

Study Completion Definition

A participant will be considered to have completed the study if the participant has completed assessments at the 1-year follow-up visit post-booster vaccination.

5. STUDY POPULATION

Cohort 1: Study sites will be provided with a list of COV3001 participants who received Ad26.COV2.S at that site and who are potentially suitable for roll-over into COV2008. Listed participants will be those currently enrolled in COV3001 (ie, have not discontinued early), on whom Day 1 and Day 29 stored serum samples are available, and without major COV3001 protocol deviations that impacted immunogenicity. In addition, suitable participants will have had their Day 29 (post primary vaccination) blood sample collected within the permitted visit window.

Cohort 2: Participants who received the 2-dose regimen of BNT162b2, either while on Pfizer study NCT04368728, or post authorization, are potentially eligible for COV2008 enrollment. All former Pfizer trial participants MUST be no longer participating in study NCT04368728, but judged by the investigator to be potentially compliant in study COV2008.

Screening suitable participants for enrollment into COV2008 will be performed within 14 days before administration of the study vaccine. Refer to Section 5.4 for conditions under which the repeat of any screening procedures are allowed.

The inclusion and exclusion criteria for enrolling participants into study COV2008 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.

For a discussion of the statistical considerations of participant selection, refer to Section 9.2.

5.1. Inclusion Criteria

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

1. Criterion modified per Amendment 3:

   1.1 **Cohort 1**: Participant received Ad26.COV2.S in COV3001. The interval between the Ad26.COV2.S primary vaccination should, preferably, be , ≥6 months prior to study vaccination on COV2008, however a window of maximum -20 days is allowed.

   Note: participants who received Ad26.COV2.S at the unblinding visit in study COV3001 will not be eligible for inclusion in this study.

   Cohort 2: Participant completed primary vaccination with a 2-dose regimen of BNT162b2 vaccine (Pfizer). The last dose of BTN162b2 should, preferably, be ≥6 months prior to study vaccination on COV2008, however a window of a maximum of -20 days is allowed.

2. Participant must provide consent indicating that he or she understands the purpose, procedures and potential risks and benefits of the study, and is willing to participate in the study.

3. Participant is willing and able to adhere to the prohibitions and restrictions specified in this protocol.

4. Participant is ≥18 years of age on the day of signing the ICF.

5. In the investigator's clinical judgment, participant may have a stable and well-controlled medical condition including comorbidities associated with an increased risk of progression to severe COVID-19 (CDC 2021a) (see Appendix 9) (including stable/well controlled HIV infection)*. If participants are on medication for a medical condition (including comorbidities associated with an increased risk of progression to severe COVID-19), the medication dose cannot have been increased within 12 weeks preceding vaccination and must be expected to remain stable for the duration of the study. Participants will be included on the basis of relevant medical history and BMI measurement at screening.

   * Stable/well-controlled HIV infection includes:

     a. Documented CD4 cell count ≥300 cells/μL within 6 months prior to screening.

b. Documented HIV viral load <50 copies/mL within 6 months prior to screening.

c. Participant must be on a stable anti-retroviral treatment (ART) for 6 months (unless the change is due to tolerability, in which case the regimen can be for only the previous 3 months; changes in formulation are allowed; nationwide guidelines that require transition from one ART regimen to another are allowed) and the participant must be willing to continue his/her ART throughout the study as directed by his/her local physician.

Note: Participants with ongoing and progressive comorbidities associated with HIV infection will be excluded but comorbidities associated with HIV infection that have been clinically stable for the past 6 months are not an exclusion criterion.

Laboratory methods for confirming a diagnosis of HIV infection are: Any evidence (historic or current) from medical records, such as ELISA with confirmation with Western Blot or real time reverse-transcriptase polymerase chain reaction (RT-PCR), or of a detectable viral load (country-specific regulatory approved tests). A laboratory result within 6 months of screening does not need to be repeated.

If a potential participant does not have HIV viral load and CD4 cell count data in his/her medical records from the last 6 months, they will be instructed to go to their local health care provider and obtain the necessary data for potential entry into the study.

8. Participant agrees to not donate bone marrow, blood, and blood products from the study vaccine administration until 3 months after receiving the study vaccine.
9. Participant must be willing to provide verifiable identification, has means to be contacted and to contact the investigator during the study.
10. Participant must be able to read, understand, and complete questionnaires in the eCOA (ie, the COVID-19 signs and symptoms surveillance question, the e-Diary, and the electronic patient-reported outcomes (ePROs) [see Appendix 1 for definition of terms])[a].

5.2. Exclusion Criteria

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

1. Participant has a clinically significant 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 planned study vaccination; randomization at a later date is permitted at the discretion of the investigator. Please notify the sponsor (or medical monitor) of this decision.

[a] Participants with visual impairment are eligible for study participation and may have caregiver assistance in completing the eCOA questionnaires.

2. Participant has a known or suspected allergy or history of anaphylaxis or other serious adverse reactions to vaccines or their excipients (including specifically the excipients of the study vaccine; refer to the IB (IB Edition 4 Ad26.COV2.S 2021 and its addenda).
3. Participant has abnormal function of the immune system resulting from:
    a. Clinical conditions (eg, autoimmune disease or potential immune mediated disease or known or suspected immunodeficiency, or participant on hemodialysis) expected to have an impact on the immune response of the study vaccine. Participants with clinical conditions stable under non-immunomodulator treatment (eg, autoimmune thyroiditis, autoimmune inflammatory rheumatic disease such as rheumatoid arthritis) may be enrolled at the discretion of the investigator. Non-immunomodulator treatment is allowed as well as steroids at a non-immunosuppressive dose or route of administration.
    b. Chronic or recurrent use of systemic corticosteroids within 6 months before administration of study vaccine and during the study. A substantially immunosuppressive steroid dose is considered to be ≥2 weeks of daily receipt of 20 mg of prednisone or equivalent.
    Note: Ocular, topical, or inhaled steroids are allowed.
    c. Administration of antineoplastic and immunomodulating agents or radiotherapy within 6 months before administration of study vaccine and during the study.
4. Participant received treatment with immunoglobulins (Ig) in the 3 months or exogenous blood products (autologous blood transfusions are not exclusionary) in the 4 months before the planned administration of the study vaccine or has any plans to receive such treatment during the study.

    Note: Specific monoclonal antibodies, not directed against SARS-CoV-2, are allowed.

5. Participant received or plans to receive:
    a. Licensed live attenuated vaccines - within 28 days before or after planned administration of study vaccine.
    b. Other licensed (not live) vaccines - within 14 days before or after planned administration of study vaccine.
6. Participant has a known history of confirmed SARS-CoV-2 infection.
7. Criterion Modified per Amendment 6

7.1 Participant received an investigational drug (including investigational drugs for prophylaxis of COVID-19) within 30 days or used an invasive investigational medical device within 30 days, received investigational Ig or monoclonal antibodies within 3 months, or received convalescent serum for COVID-19 treatment within 4 months, or received an investigational vaccine (excluding Ad26.COV2.S or BNT162b2) within 6 months before the planned administration of the study vaccine or is currently enrolled or plans to participate in another investigational study during the course of this study. See also Section 6.7.

Note: Participation in an observational clinical study is allowed at the investigator's discretion; please notify the sponsor (or medical monitor) of this decision.

8. Participant has a history of an underlying clinically significant acute or 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.
9. Participant has a history of heparin-induced thrombocytopenia or thrombosis in combination with thrombocytopenia.
10. Participant has a contraindication to IM injections and blood draws eg, bleeding disorders.
11. Participant has, or has had, major psychiatric illness which in the investigator's opinion would compromise the participant's safety or compliance with the study procedures.
12. Participant cannot communicate reliably with the investigator.
13. Participant who, in the opinion of the investigator, is unlikely to adhere to the requirements of the study, or is unlikely to complete the full course of vaccination and observation.
14. Participant has a history of acute polyneuropathy (eg. Guillain-Barré syndrome).
15. Employee of the investigator or study site who has direct involvement in the proposed study.
16. History of capillary leak syndrome.

NOTE: Investigators should ensure that all study enrollment criteria have been met prior to vaccination. If a participant's clinical status changes (including any available laboratory results or receipt of additional medical records) after screening but before the study vaccine is given such that the participant no longer meets all eligibility criteria, then the participant should be excluded from participation in the study.

5.3. Lifestyle Considerations

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

1. Refer to Section 6.7 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.
3. Agree to follow requirements for the electronic completion of the COVID-19 signs and symptoms surveillance question in the eCOA.

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.

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened once. Participants who are rescreened will be assigned a new participant number, undergo the ICF process, and then restart a new screening phase.

5.5. Criteria for Temporarily Delaying Administration of Study Vaccine

The following events constitute a temporary contraindication to study vaccination:

- Clinically significant acute illness at the time of vaccination. This does not include minor illnesses, such as diarrhea or mild upper respiratory tract infection.
- Fever (body temperature ≥38.0°C/100.4°F) within 24 hours prior to the planned time of vaccination.

If any of these events occur at the scheduled time for vaccination, randomization at a later date within the screening window is permitted at the discretion of the investigator and after consultation with the sponsor. If randomization cannot occur within the screening window, rescreening is required. Vaccination can be rescheduled, as long as this is in agreement with the allowed windows. If the vaccination visit cannot be rescheduled within the allowed window or the contraindications to vaccination persist, the sponsor should be contacted for further guidance.

6. STUDY VACCINATION AND CONCOMITANT THERAPY

6.1. Study Vaccine Administration

Ad26.COV2.S will be supplied at a concentration of 1×10^{11} vp/mL, as suspensions in single-use vials. The Ad26.COV2.S vaccine will be dosed at 5×10^{10} vp, 2.5×10^{10} vp and 1×10^{10} vp. Formulation buffer (diluent) will be supplied as 15 mM citrate, 5% (w/w) hydroxypropyl-β-cyclodextrin, 0.4% (w/w) ethanol, 0.03% (w/w) polysorbate 80, 75 mM NaCl, pH 6.2. Formulation buffer is withdrawn from one vial and added to a vial of vaccine to achieve the required vp concentrations for different dose levels (full details in IPPI). Then 0.5 mL is withdrawn from the vial for injection.

Participants will be vaccinated with the study vaccine on Day 1:

Study vaccines will be administered by IM injection into the deltoid muscle, preferably of the non-dominant arm. If an injection cannot be given in the deltoids due to a medical or other contraindication (for example, tattooed upper arms rendering it difficult to assess site reactogenicity), use alternative locations such as the hip, thigh or buttocks (to be avoided in overweight participants). In all circumstances, IM injections in other locations than the upper arm are not considered protocol deviations.

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

Study vaccines will be manufactured and provided under the responsibility of the sponsor. Refer to the IB for a list of excipients (IB Edition 4 Ad26.COV2.S 2021 and its addenda).

Refer to the study site investigational product and procedures manual (SIPPM) and the Investigational Product Preparation Instructions (IPPI) for additional guidance on study vaccine administration.

6.2. Preparation/Handling/Storage/Accountability

Preparation/Handling/Storage

All study vaccine must be stored in a secured location with no access for unauthorized personnel and at controlled temperatures as indicated on the clinical labels. If study vaccine is exposed to temperatures outside the specified temperature range, all relevant data will be sent to the sponsor to determine if the affected supplies can be used or will be replaced. The affected study vaccine must be quarantined and not used until further instruction from the sponsor is received.

Refer to the SIPPM and the IPPI for additional guidance on study vaccine preparation, handling, and storage.

An unblinded study-site pharmacist, or other qualified individual, who will have no other study function following vaccination, will prepare the appropriate vials and syringes, labeled with the participant's identification number, and provide the syringes for the study vaccine in a blinded manner to the blinded vaccine administrator (a trained and qualified study nurse, medical doctor, or otherwise qualified health care professional [HCP]) who will perform the injection.

Accountability

The investigator is responsible for ensuring that all study vaccines received at the site are inventoried and accounted for throughout the study. The study vaccines 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 vaccines 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 vaccines 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 vaccines will be documented on the vaccine accountability 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 accountability form.

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

Study vaccines should be dispensed under the supervision of the investigator or a qualified member of the study site personnel, or by a hospital/clinic pharmacist. Study vaccines will be supplied only to participants participating in the study. Returned study vaccines must not be dispensed again, even to the same participant. Study vaccines may not be relabeled or reassigned for use by other participants. The investigator agrees neither to dispense the study vaccines from, nor store it at, any site other than the study sites agreed upon with the sponsor. Further guidance and information for the final disposition of unused study vaccines are provided in the SIPPM.

6.3. Measures to Minimize Bias: Randomization and Blinding

Vaccine Allocation

Procedures for Randomization

Central randomization will be implemented in this study. Cohort 1 participants will be randomly assigned to 1 of 3 Ad26.COV2.S vaccine groups (5×10^{10} vp [Group 1], 2.5×10^{10} vp [Group 2] and 1×10^{10} vp [Group 3]).

Cohort 2 participants will be randomly assigned to 1 of 3 Ad26.COV2.S vaccine groups (5×10^{10} vp [Group 4] 2.5×10^{10} vp [Group 5] and 1×10^{10} vp [Group 6] see Section 4.1).

Randomization will be based on a computer-generated randomization schedule prepared before the study by, or under the supervision of, the sponsor. The randomization will be balanced by using randomly permuted blocks. The interactive web response system (IWRS) will assign a unique intervention code, which will dictate the intervention assignment and matching study intervention kit for the participant. 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.

If, due to the urgency of study initiation during the ongoing pandemic, the IWRS is not yet available at the planned time of randomization of the first participant, randomization may be started based on a paper randomization list until the IWRS is live.

The study will be stratified for age using 2 age strata: 18-59 and ≥60 years of age.

Blinding

The investigator will not be provided with randomization codes. IWRS 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 vaccine assignment (ie, immunogenicity data, study vaccine accountability data, study vaccine allocation, biomarker or other specific laboratory data) will be

handled with special care to ensure that the integrity of the blind is maintained and the potential for bias is minimized. This can include making special provisions, such as segregating the data in question from view by the investigators, clinical team, or others as appropriate.

Under normal circumstances, the blind should not be broken until the database is finalized, with two exceptions. First, the investigator may, in an emergency, determine the identity of the intervention by contacting the IWRS. While the responsibility to break the intervention code in emergency situations resides solely with the investigator, 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, time, and reason for the unblinding must be documented by the IWRS and in the source document.

At selected study sites, the sponsor may unblind participants who are potentially eligible to roll over into sponsor-supported study VAC31518COV2015. This unblinding of selected participants may occur after all participants at the selected site have completed their 6-month study visit procedures.

When possible, participants who have had their vaccine assignment unblinded should continue to return for scheduled evaluations unless they have been selected and enrolled into VAC31518COV2015, in which case the participants will discontinue the Schedule of Activities for the current study and begin the Schedule of Activities for VAC31518COV2015. In this event, participants will have an early exit visit for COV2008 prior to enrolling in VAC31518COV2015.

In general, randomization codes will be disclosed fully only if the study is completed and the clinical database is closed. However, if a primary analysis is specified, the randomization codes and, if required, the translation of randomization codes into intervention and control groups will be disclosed to those authorized and only for those participants included in the primary analysis.

If randomized participants are withdrawn from vaccination before the study vaccine is administered, additional participants may be recruited to replace these participants at the discretion of the sponsor. Any replacement participant will be assigned to the same group as the original (discontinued) participant. If randomized participants are withdrawn after the study vaccine is administered, they will not be replaced.

In the event that randomization is started based on a paper randomization list, sealed randomization codes will be provided for each participant containing coded details of study vaccine allocation. All randomization codes, whether opened or sealed, will be collected after the end of the participant's participation in the study. If emergency unblinding is required, the investigator may determine the identity of the study vaccine by opening the sealed code. The date, time, and reason for the unblinding must be documented in the appropriate section of the eCRF, and in the source document.

6.4. Study Vaccination Compliance

Study vaccines will be administered IM by blinded qualified study site personnel at the study site. The date and time of study vaccine administration and the location used will be recorded in the eCRF.

6.5. Dose Modification

Dose modification is not applicable in this study.

6.6. Treatment of Overdose

For this study, any dose of Ad26.COV2.S greater than the highest dose tested in this study will be considered an overdose. The sponsor does not recommend specific treatment 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 AEs/AESIs/SAEs (ie, the participant will remain at the study site for at least 1 hour and will be closely monitored for allergic or other reaction by study staff. Follow-up telephone calls 12 hours and 24 hours post-dose will be made).
- Document the quantity of the excess dose in the eCRF.
- Report as a special reporting situation (see Appendix 10.4.4).

6.7. Concomitant Therapy

Prestudy specific therapies such as analgesic/antipyretic medications and non-steroidal anti-inflammatory drugs, corticosteroids, antihistamines, and vaccinations administered up to 30 days before study vaccination must be recorded at screening.

Concomitant therapies such as analgesic/antipyretic medications and non-steroidal anti-inflammatory drugs, corticosteroids, antihistamines, and vaccinations must be recorded until 28 days after administration of study vaccine. All other concomitant therapies should also be recorded if administered in conjunction with a confirmed COVID-19 case or new or worsening AEs reported per protocol requirements outlined in Section 8.4.1.

For all participants, concomitant therapies associated with an SAE or suspected AESI meeting the criteria outlined in Sections 10.4.1 and 8.4.7, respectively, will be collected and recorded in the eCRF from the moment of vaccination through the end of the study.

For all participants, concomitant therapies associated with unsolicited AEs will be collected and recorded in the eCRF from the time of vaccination through 28 days after vaccination. Concomitant therapies associated with solicited AEs will be collected by the participants and recorded in the eCRF from the time of vaccination through 7 days after vaccination.

Use of any experimental medication (including experimental vaccines other than the study vaccines) during the study is not allowed. Any participant who has received an anti-COVID-19

vaccine (other than the study vaccine or Pfizer BNT162b2) or treatment will not receive study vaccination. Participants may not receive an investigational drug (including investigational drugs for prophylaxis of COVID-19) or use an invasive investigational medical device within 30 days or receive an investigational Ig or monoclonal antibodies within 3 months, or receive convalescent serum for COVID-19 treatment within 4 months or receive an investigational vaccine (including investigational Adenoviral-vectored vaccines other than Ad26.COV2.S) within 6 months before the planned administration of study vaccine. Cohort 2 participants will be required to provide the dates they received primary vaccination with BNT162b2. Investigators are required to enter this information in the eCRF.

Licensed live attenuated vaccines should be given at least 28 days before or at least 28 days after a study vaccination. Other licensed (not live) vaccines (eg, tetanus, hepatitis A, hepatitis B, rabies) 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. The use of any coronavirus vaccine (licensed/authorized or investigational) other than Ad26.COV2.S and BNT162b2, is disallowed at any time prior to vaccination and during the study. If a vaccine is indicated in a post-exposure setting (eg, rabies or tetanus), it must take priority over the study vaccine.

Antipyretics are recommended post-vaccination for symptom relief as needed. Prophylactic antipyretic use is not encouraged; however, in some instances, it could be considered for participants with special circumstances and/or comorbidities.

Chronic or recurrent use of systemic corticosteroids[a], at immunosuppressive dose and administration of antineoplastic and immunomodulating agents or radiotherapy is prohibited during the study and within 6 months before the planned administration of the study vaccine. If any of these agents are indicated in a disease setting, these must take priority over the study vaccine. Refer to Section 5.2 for further details of prohibited therapy.

The sponsor must be notified in advance (or as soon as possible thereafter) of any instances in which prohibited therapies are administered. Depending on the time of the occurrence, any participant who receives a prohibited concomitant medication will not be included in the immunogenicity analyses.

6.8. Study Pausing Rules

The sponsor (including designated sponsor teams) and/or Sponsor Committee as well as the investigator(s) will monitor safety in a blinded manner. Adverse events that may lead to the study vaccination pausing rules are described below and will be assessed by the designated sponsor team/committee to confirm that the study pause is warranted.

The occurrence of any of the following events will lead to a pause in further study vaccination:

[a] Note: Ocular, topical or inhaled steroids are allowed.

1. Death of a participant, considered related to study vaccine or if the causal relationship to the study vaccine cannot be excluded; OR
2. One or more participants experience an SAE (solicited or unsolicited) that is determined to be related to study vaccine; OR
3. One or more participants experience anaphylaxis or generalized urticaria, clearly not attributable to other causes than vaccination with study vaccine

To enable prompt response to a situation that could trigger pausing rules, the investigator should notify the sponsor's medical monitor or designee (AND email the SAE form to Global Medical Safety Operations, if applicable), immediately and no later than 24 hours after becoming aware of any related SAE AND update the eCRF with relevant information on the same day the SAE information is collected (see also Section 8.4). Based on the pausing criteria, the sponsor's medical monitor or designee then decides whether a study pause is warranted and informs the IDMC of the decision. All sites will be notified immediately in case of a study pause. The sponsor's medical monitor or designee is responsible for the immediate notification of IDMC members and coordination of a IDMC meeting in case of a study pause.

In the case of a study pause, the IDMC will initially review blinded data, but can request unblinded data, and will make recommendations regarding the continuation of the study to the sponsor study team. Resumption of vaccinations will start only upon receipt of written recommendations by the IDMC. The clinical site(s) will be allowed to resume activities upon receipt of a written notification from the sponsor. The formal recommendation from the IDMC will be forwarded by the investigator to the IRB and by the sponsor to the relevant health authorities, according to local standards and regulations.

Vaccinations for an individual participant may be suspended for safety concerns other than those described in the pausing criteria, at the discretion of the investigator if he/she feels the participant's safety may be threatened. The sponsor's medical monitor or designee or the investigator(s) (upon consultation with the sponsor's medical monitor or designee) may initiate IDMC review for any single event or combination of multiple events which, in their professional opinion, could jeopardize the safety of the participants or the reliability of the data.

Vaccinations for the study may be suspended for safety concerns other than those described above, or before pausing rules are met, if, in the judgment of the IDMC, participant safety may be threatened.

7. DISCONTINUATION OF STUDY VACCINATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1. Discontinuation of Study Vaccination

Not applicable.

7.2. Participant Discontinuation/Withdrawal From the Study

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

- Lost to follow-up
- Withdrawal of consent
- Death
- Repeated failure to comply with protocol requirements

When a participant withdraws before study completion, the reason for withdrawal is to be documented in the eCRF and in the source document. If the reason for withdrawal from the study is withdrawal of consent, lost to follow-up, or death, then no additional assessments are allowed.

Any participants who receive a COVID-19 booster vaccine off study will remain on study (continuing with all scheduled procedures and follow-up) but will be excluded from the formal hypothesis testing of the primary objectives. Investigators are required to enter the off-study booster information in the eCRF.

Withdrawal of Consent

A participant declining to return for scheduled visits does not necessarily constitute withdrawal of consent. Alternate follow-up mechanisms that the participant agreed to when signing the consent form apply as local regulations permit.

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 10.3.5. 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

To reduce the chances of a participant being deemed lost to follow-up, prior to randomization attempts should be made to obtain contact information from each participant, eg, home, work, and mobile telephone numbers and email addresses for both the participant as well as appropriate family members.

A participant will be considered lost to follow-up if the participant repeatedly fails to return for scheduled visits and is unable to be contacted by the study site. A participant cannot be deemed lost to follow-up until all reasonable efforts made by the study site personnel to contact the participant are deemed futile. The following actions must be taken if a participant fails to return to the study site for a required study visit:

- The study site personnel must attempt to contact the participant to reschedule the missed visit as soon as possible, to counsel the participant on the importance of maintaining the assigned visit schedule, to ascertain whether the participant wishes to or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee must make every reasonable effort to regain contact with the participant (where possible, 3 telephone calls,

emails, fax, and, if necessary, a certified letter to the participant's last known mailing address, or local equivalent methods). Locator agencies may also be used as local regulations permit. These contact attempts should be documented in the participant's medical records.

- Should the participant continue to be unreachable, they will be considered to have withdrawn from the study.

Should a study site close, eg, for operational, financial, or other reasons, and the investigator cannot reach the participant to inform them, their contact information will be transferred to another study site.

8. STUDY ASSESSMENTS AND PROCEDURES

Overview

The Schedule of Activities summarizes the frequency and timing of all measurements applicable to this study.

All participants will have access to an eCOA digital tool. This eCOA will be used to collect COVID-19 signs and symptoms surveillance info for all participants, ePRO (Symptoms of infection with Coronavirus-19 [SIC], including body temperature, and pulse oximetry results) for all participants at baseline and in case of COVID-19-like signs and symptoms, and e-Diary data on 7-day reactogenicity (solicited signs and symptoms, including body temperature). All eCOA assessments should be conducted/completed before any tests, procedures, or other consultations to prevent influencing participant responses. Refer to the PRO completion guidelines for instructions on the administration of ePROs.

If multiple assessments are scheduled for the same timepoint, it is recommended that procedures be performed in the following sequence: vital signs before blood draws. If needed, assessments may be performed at another day within the applicable visit window. Actual dates and times of assessments will be recorded in the source document and in the eCRF.

All participants will be provided a thermometer to measure body temperature. Participants will be provided a ruler (to measure local injection site reactions) and a participant e-Diary in the eCOA digital tool to record body temperature and solicited local (at injection site) and systemic signs and symptoms. The e-Diary includes instructions on how to capture the data and grading scales to assess severity of the signs and symptoms post-vaccination (reactogenicity). The study staff is responsible for providing appropriate training to the participant to avoid missing or incorrect data. The e-Diary will be reviewed by the study personnel at visits indicated in the Schedule of Activities. If the e-Diary review is missed, the diary will be reviewed during the following visit.

All participants will also be provided with a kit to collect nasal swabs samples and receptacles to collect saliva (see Section 8.1.2).

Over the course of the study, a total approximate blood volume up to a maximum of 180 mL will be collected in participants not included in any of the subsets. A total approximate blood volume up to a maximum of 405 mL will be collected in a subset of participants (Subset 4), which will

include safety and cellular immunogenicity assessments on PBMC (if feasible). For the participants included in subsets 1, 2, and 3, a total blood volume of approximately 195 mL will be collected for safety and immunogenicity assessments without PBMC. For participants enrolled at the BIDMC site, up to an additional 50 mL of blood per blood draw will be collected for experimental research amounting to an additional total approximate blood volume of 300 mL. For participants with a suspected AESI, additional samples up to approximately 30 mL of blood will be collected. Additional blood samples (up to 35 mL) will be collected from participants that experience COVID-19-like signs and symptoms meeting prespecified criteria for suspected COVID-19. The total blood volume to be collected is within the US Department of Health and Human Services Office for Human Research Protections, and US Food and Drug Administration (FDA) guidelines of 550 mL in any 8-week period. Refer to the Schedule of Activities for the timings of blood samples over the course of the study, and in the event of a suspected COVID-19 episode. Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

Study visits, other than screening and visits at which study vaccination is scheduled, may take place at a remote lab service or the participant's home if there are travel restrictions in case of an ongoing pandemic.

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 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.

Visit Windows

Visit windows are provided in the Schedule of Activities in Section 1.3. The participant should be encouraged to come on the exact day planned and resort to use the visit window only when absolutely necessary.

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

Screening

Screening will be performed within 14 days prior to study 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 and the predose assessment does not need to be performed. Screening must be completed and all eligibility criteria must be fulfilled prior to randomization and vaccination.

Screening may be conducted in part via a sponsor- and IRB-pre-approved non-study-specific screening consent process, but only if the relevant pre-screening tests are identical to the per protocol screening tests and are within 2 weeks prior to vaccination. However, no study-specific procedures, other than these pre-approved pre-screening assessments, will be performed until the participant has signed the study-specific ICF. The study-specific ICF date will be entered into the eCRF. The non-study-specific ICF will be considered source data.

Study-Specific Materials

The investigator will be supplied with the following:

- IB for Ad26.COV2.S
- Package Insert for Ad26.COV2.S
- Thermometer
- Ruler (to measure diameter of any erythema and swelling)
- A pulse oximeter
- Pharmacy manual (IPPI)/SIPPM
- Laboratory manual
- IWRS manual
- eCRF completion guidelines
- Sample ICF
- Nasal swab kits, saliva recipients, and participant instructions
- eCOA platform access and participant instructions. Participants may use their own eDevice using an application if their device (smartphone or tablet) is compatible, or a web portal. Provisioned devices will be available on a limited basis.
- Lab kits/blood collection tubes and aliquoting materials
- PBMC binders
- ePRO completion instructions
- Participant diaries
- Contact information page(s)

8.1. COVID-19 and Immunogenicity Assessments

8.1.1. Prespecified Criteria for Suspected COVID-19

The criteria for suspected COVID-19 (ie, the triggers to proceed with home-collection of the nasal swabs on COVID-19 Day 1-2 and to proceed with the COVID-19 Day 3-5 visit) are prespecified as follows:

- **A positive RT-PCR result for SARS-CoV-2, through a private or public laboratory independent of the study, whether symptomatic or asymptomatic**

  OR

- **New onset or worsening of any 1 of these symptoms, which lasts for at least 24 hours, not otherwise explained:**
  - Headache
  - Malaise (appetite loss, generally unwell, fatigue, physical weakness)
  - Myalgia (muscle pain)
  - Chest congestion
  - Cough
  - Runny nose
  - Shortness of breath or difficulty breathing (resting or on exertion)
  - Sore throat
  - Wheezing
  - Eye irritation or discharge
  - Chills
  - Fever (≥38.0°C or ≥100.4°F)
  - Pulse oximetry value ≤95%, which is a decrease from baseline
  - Heart rate ≥90 beats/minute at rest, which is an increase from baseline
  - Gastrointestinal symptoms (diarrhea, vomiting, nausea, abdominal pain)
  - Neurologic symptoms (numbness, difficulty forming or understanding speech)
  - Red or bruised looking toes
  - Skin rash
  - Taste loss or new/changing sense of smell
  - Symptoms of blood clots: pain/cramping, swelling or redness in your legs/calves
  - Confusion

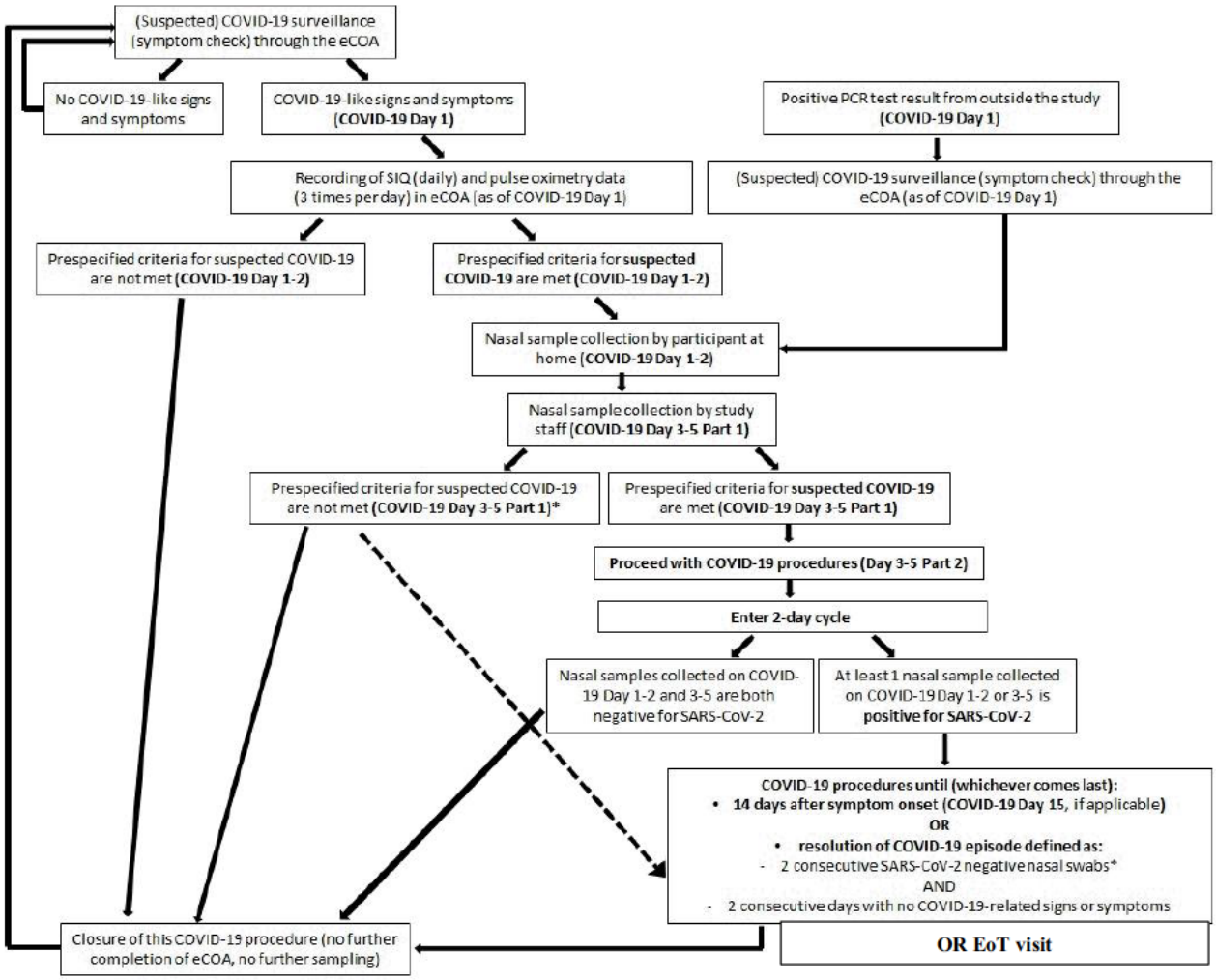
- Bluish lips or face
- Clinical suspicion/judgment by investigator of symptoms suggestive for COVID19

As several of the prespecified criteria for suspected COVID-19 overlap with vaccine-related reactogenicity, investigators' clinical judgment is required to exclude vaccine-related events when assessing suspected COVID-19.

8.1.2. Procedures in the Event of COVID-19-like Signs and Symptoms

Procedures to be performed in the event a participant experiences signs or symptoms suggesting possible COVID-19 are detailed in the Schedule of Activities in Section 1.3.3. A high-level schematic overview is presented in Figure 2.

Figure 2: Decision Tree for COVID-19 Procedures



If the participant no longer meets the prespecified criteria at Day 3-5 and results from the nasal sample at Day 1-2 and/or Day 3-5 are latently positive (i.e. ≥14 days to result), the participant will be contacted and asked to proceed with COVID-19 procedures (2-day cycles).

If signs and symptoms are still ongoing on **COVID-19 Day 3-5**, collection of SIC will be continued until at least 14 days after onset unless both **COVID-19 Day 1-2** and **COVID-19 Day 3-5** are both negative. If either of the swabs is positive or the result is unknown AND the participant is beyond 14 days after onset of symptoms, the SIC can be stopped after 2 days without signs and symptoms.

*If 2 consecutive nasal swabs negative for SARS-CoV-2 are not available due to operational reasons (eg, delays in results availability), participants may cease collection of nasal swabs and saliva samples after COVID-19 Day 29, provided they have 2 consecutive days with no COVID-19-related signs and symptoms. In these cases, participants may be asked to resume sample collection if nasal sample results—once available—do not present with 2 consecutive negative swabs for SARS-CoV-2.

COVID-19 = coronavirus disease-2019; eCOA = electronic clinical outcome assessment; EoT= end of trial; PCR= polymerase chain reaction; SARS-CoV-2 = severe acute respiratory syndrome coronavirus-2; SIC = Symptoms of Infection with Coronavirus-19

For all medical visits for COVID-19 or COVID-19 complications, including those resulting in hospitalization, a standard list of questions will be provided (MA-COV form [Appendix 7]), with the aim to collect additional information on any other diagnostics (eg, chest X-rays, spirometry, pulmonary function tests) or interventions during the clinical course of COVID-19. The MA-COV form will be provided to the participant at the vaccination visit and should be completed by the medical care provider or the study site personnel during medical visits for COVID-19 or COVID-19 complications.

Note: if for any reason a site visit per the procedures described below is not feasible, a member of the study staff can visit the participant at home (or at the hospital or other location, if needed), if allowed by local regulations.

Day 1-2 procedures in case of signs and symptoms

If a participant (or their designated caregiver) records in the eCOA or informs the site that he/she experienced any signs or symptoms suggesting possible COVID-19, this will be considered **COVID-19 Day 1** (day of onset of signs and symptoms). The participant will be asked to complete the ePROs (ie, the SIC [Appendix 5], including body temperature) in the eCOA.

Notes:

The SIC questionnaire asks the participant if he/she had any of the prespecified signs or symptoms (see Appendix 5) during the past 24 hours, and (when applicable) to rate the severity. The SIC questionnaire takes approximately 5 minutes to complete.

The participant should record the highest temperature in the last 24 hours in the SIC.

The participant should record at least 1 of the 3 pulse oximetry readings in the last 24 hours in the eCOA.

If a participant is unable to complete the SIC in the eCOA, a study staff member can collect information on the participant's symptoms and body temperature, by contacting the participant by telephone (or visit the participant at home), reading the questions aloud to the participant and entering the participant's responses on the participant's behalf. More details are provided in eCOA Study Manual.

If a participant is unable to complete the SIC in eCOA, the reason for missing the SIC completion should be recorded in the eCRF.

Based on the information collected through the SIC, the site will reach out to the participant at the latest on COVID-19 Day 2 (the day after the day of symptom onset) to assess whether the reported signs and symptoms qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1). As several of the prespecified criteria for suspected COVID-19 overlap with vaccine-related reactogenicity, investigators' clinical judgment is required to exclude vaccine-related events when assessing suspected COVID-19. If the participant would actively reach out to the site already on COVID-19 Day 1, the site should already make a first assessment on COVID-19 Day 1 to check whether the reported signs and symptoms qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1). As soon as the prespecified criteria for

suspected COVID-19 are met (**COVID-19 Day 1-2**), the participant will be asked to undertake the COVID-19 procedures. In particular:

The participant will be asked to continue to complete the ePROs in the eCOA as specified above for COVID-19 Day 1:

- SIC (including body temperature): every day, preferably in the evening around the same time each day.
- Blood oxygen saturation and pulse rate using a pulse oximeter 3 times a day, preferably in the morning, at lunch time, and in the evening.

  Note: the ePROs do not have to be completed if special circumstances occur, such as hospitalization or ventilation, in which case the reason for not completing the ePROs should be recorded by site staff in the eCRF.

The participant will be asked to collect a nasal swab at home on **COVID-19 Day 1-2**, as soon as possible after it has been confirmed that the prespecified criteria for suspected COVID-19 are met. If the participant requires assistance, a trained HCP can help the participant to collect the nasal swab. The study site should arrange transfer of the nasal swab to the study site as soon as possible after collection, preferably within 24 hours. The COVID-19 Day 1-2 nasal swab can also be collected at the study site (or hospital or other location, if needed), if preferred by the participant.

Day 1-2 procedures in case of a positive RT-PCR test outside the study site context

If a participant becomes aware of a positive RT-PCR test for SARS-CoV-2 he/she should contact the site as soon as possible. The day the participant became aware of the positive RT-PCR test will be considered **COVID-19 Day 1**. Regardless of whether the participant is symptomatic or asymptomatic, they will be asked to:

Complete the (suspected) COVID-19 surveillance (symptom check) in the eCOA. In case of COVID-like signs and symptoms they will need to complete the SIC (Appendix 5) including body temperature) in the eCOA.

The participant will be asked to collect a nasal swab at home on **COVID-19 Day 1-2**, as described for the participants with signs and symptoms (see above).

These precautionary measures are to ensure that site staff who come into physical contact with a participant deemed to be a COVID-19 case undertake the proper safety procedures such as wearing of personal protective equipment.

Day 3-5 procedures for all participants who have met the prespecified criteria for (suspected) COVID-19

The participant will be asked to come to the site on **COVID-19 Day 3-5** (between 2 and 4 days after symptom onset/becoming aware of a positive RT-PCR test).

If a site visit is not feasible, a member of the study staff or designee could visit the participant at home (or at the hospital or other location, if needed), if allowed by local regulations. The study staff or designee visiting participants at home will use personal

protective equipment according to local regulations. The COVID-19 Day 3-5 assessments may also be performed by a trained HCP, if allowed per local regulations.

During **Part 1** of the **COVID-19 Day 3-5** visit, if the participant has experienced COVID-19 like signs and symptoms, the site will interview the participant to assess whether the reported signs and symptoms still qualify as a suspected COVID-19 episode using prespecified criteria (Section 8.1.1). In addition, for all participants with (suspected) COVID-19, a qualified member of the study site will measure vital signs (body temperature, blood pressure, heart rate, and respiratory rate) and pulse oximetry. A targeted physical examination will be performed based on the judgment of the investigator. A nasal swab will be collected for detection of SARS-CoV-2 by a qualified member of the study site.

If the signs and symptoms still meet the prespecified criteria for suspected COVID-19 on COVID-19 Day 3-5 or if at least one nasal sample from COVID-19 Day 1-2 or Day 3-5 visits is positive for SARS-CoV-2 (tested by RT-PCR), the following assessments and procedures are to be performed during **Part 2** of the **COVID-19 Day 3-5** visit: a blood sample for exploration of biomarkers that correlate with SARS-CoV-2 infection and COVID-19 severity will be collected by a qualified member of the study site. A saliva sample will be taken by the participant during the study visit. The MRU questionnaire will be completed based on a clinical interview (Appendix 6). The medical history and description of COVID-19 episode will be collected by interview with the participant.

If signs and symptoms are still ongoing on **COVID-19 Day 3-5**, collection of SIC will continue as specified in the next section (***Closure of the COVID-19 episode***) ***Closure of the COVID-19 episode***

If the signs and symptoms no longer meet the prespecified criteria for suspected COVID-19 on COVID-19 Day 3-5 and no result from nasal swabs collected on Day 1-2 and/or Day 3-5 visits is available, the participant will not undertake any further COVID-19 procedures. He/she will fall back to the default Schedule of Activities, until the end of the study/early withdrawal.

Procedures during the 2-day cycles

If a participant has signs and symptoms that still meet the prespecified criteria for suspected COVID-19 (Section 8.1.1) at COVID-19 Day 3-5 visit or has at least one positive nasal sample for SARS-CoV-2 at COVID-19 Day 1-2 or COVID-19 Day 3-5 visits, he or she will be asked to undertake the COVID-19 procedures, in particular:

All participants will be asked to collect a nasal swab and a saliva sample at home once every 2 days (daily alternating between nasal swabs and saliva samples). If the participant requires assistance, a trained HCP can help the participant to collect the nasal swabs and/or saliva samples. The study site should arrange transfer of the nasal swabs and saliva samples to the study site within 3 days after collection. Details are provided in the laboratory manual.

In case of signs and symptoms: The participant will be reminded to further complete the ePROs in the eCOA as described for COVID Day 1-2:

In case the nasal swabs collected on Day 1-2 or Day 3-5 visits are tested positive for SARS-CoV-2 and the participant is asymptomatic: The participant will be reminded to further complete (suspected) COVID-19 surveillance (symptom check).

If, on COVID-19 Day 3-5, the participant stopped the COVID-19 procedures and returned to default Schedule of Activities, due to lack of signs and symptoms and unavailability of results from nasal swabs collected on Day 1-2 and/or Day 35 visits, the participant will be contacted as soon as at least one of these samples is found to be positive for SARS-CoV-2 presence. The participant will be asked to resume COVID-19 procedures, until 14 days after symptom onset (COVID-19 Day 15) or until **resolution of the COVID-19 episode**, whichever comes last.

Notes:

- Participants should be encouraged by the site to collect nasal swabs and saliva samples as indicated in the Schedule of Activities (Section 1.3). If the participant is unable or unwilling to collect all samples as requested, the participant should still complete the other COVID19 assessments, including the visit at COVID-19 Day 29.

Day 29 procedures

If a participant has at least 1 SARS-CoV-2 positive nasal swab collected on COVID-19 Day 1-2 or Day 3-5, then he or she will be asked to return to the site on COVID-19 Day 29 (±7 days) where a blood sample will be drawn for sero-confirmation and exploration of biomarkers that correlate with SARS-CoV-2 infection and COVID-19 severity. A qualified member of the study site will measure vital signs (body temperature, blood pressure, heart rate, and respiratory rate) and pulse oximetry. A targeted physical examination will be performed based on the judgment of the investigator. The MRU questionnaire will be completed based on a clinical interview (Appendix 6). The medical history and description of COVID-19 episode will be collected by interview with the participant. If the participant is still symptomatic, he/she will complete the SIC (Appendix 5) in the eCOA. Asymptomatic participants will complete the (suspected) COVID-19 surveillance (symptom check).

Notes: COVID-19 Day 29 should still be performed even if the nasal swabs results are still pending. The COVID-19 Day 29 assessments may also be performed by a trained HCP at the participant's home, if allowed per local regulations.

This visit can be combined with a regular study visit if within the applicable visit windows.

Closure of the COVID-19 episode

The participant should continue the COVID-19 procedures until any of the following occurs, based on molecular test results:

If both nasal swabs (collected on COVID-19 Day 1-2 and COVID-19 Day 3-5) are **negative** for SARS-CoV-2, the participant will not undertake any further COVID-19 procedures and will fall back to the default Schedule of Activities, until the end of the study/early withdrawal.

If the participant has at least 1 SARS-CoV-2 positive nasal swab collected on COVID-19 Day 1-2 or Day 3-5 visits, then the participant will be asked to undertake the COVID-19

procedures (2-day cycles) until the EoT visit or 14 days after symptom onset (COVID-19 Day 15) or until **resolution of the COVID-19 episode**, whichever comes last[a]. Resolution of the COVID-19 episode is defined as having 2 consecutive SARS-CoV-2 negative nasal swabs and 2 consecutive days with no COVID-19-related signs or symptoms. Once past COVID-19 Day 15, participants should stop the collection of nasal swabs and saliva samples as soon as 2 consecutive nasal swabs are SARS-CoV-2 negative, but (if still symptomatic at that time) should continue completing the ePROs (including SIC, body temperature, and pulse oximetry) in the eCOA until 2 consecutive days with no COVID-19-related signs or symptoms.

Note: for participants who have signs and symptoms present at baseline (assessed pre-vaccination), only signs and symptoms that are associated with COVID-19 and that developed during the COVID-19 episode are to be taken into account.

Note: For participants experiencing COVID-19 episodes close to the end of their study participation, COVID-19 episode procedures will be followed up to their EoT visit e.g. if an episode starts -4 days prior to a scheduled EoT visit, COVID-19 Day 1- 2 and COVID-19 Day 3-5 procedures will be followed as much as possible, and then study procedures will be stopped.

If signs and symptoms are still ongoing on **COVID-19 Day 3-5**, collection of SIC will be continued until at least 14 days after onset unless both **COVID-19 Day 1-2** and **COVID-19 Day 3-5** are both negative. If either of the swabs is positive or the result is unknown AND the participant is beyond 14 days after onset of symptoms, the SIC can be stopped after 2 days without signs and symptoms.

If 2 consecutive nasal swabs negative for SARS-CoV-2 are not available due to operational reasons (eg, delays in results availability), participants may cease collection of nasal swabs and saliva samples after COVID-19 Day 29, provided they have 2 consecutive days with no COVID-19-related signs and symptoms. In these cases, participants may be asked to resume sample collection if nasal sample results once available do not present with 2 consecutive negative swabs for SARS-CoV-2.

Upon closure of the COVID-19 episode and procedures, all participants will fall back to the default Schedule of Activities (Section 1.3), until the end of the study/early withdrawal. All confirmed COVID-19 episodes will be communicated to the respective participant and to other authorities according to local regulations.

If the participant experiences new signs or symptoms suggesting possible COVID-19 at a later point in time, the participant would re-start the COVID-19 procedures from COVID-19 Day 1 onwards.

With regards to the ePRO (ie, the SIC, including body temperature):

- The ePRO instrument will be provided in the local language in accordance with local guidelines.

[a] long-term sequelae of COVID-19 will not be followed until their resolution if not resolved within a month.

- The ePRO instrument must be available for regulators and for IRB/ERC submissions, therefore the ePRO instrument or screen shots need to be attached to the protocol or provided in a companion manual with the instruments that will be submitted with the protocol.
- The ePRO and AE data will not be reconciled with 1 another.

8.1.3. Monitoring for Asymptomatic SARS-CoV-2 Infection

Analysis of antibodies binding to the SARS-CoV-2 N protein will be measured at Day 1 by non-S protein assays (eg, N protein ELISA and/or SARS-CoV-2 immunoglobulin assay that is dependent on the SARS-CoV-2 N protein). Participants who are SARS-CoV-2 N protein positive at any time during the study will be excluded from the immunogenicity analysis from that timepoint onwards (see Section 9.3).

8.1.4. Clinical Severity Adjudication Committee

The Clinical Severity Adjudication Committee will be utilized for adjudication of the severity of COVID-19 cases taking into account all available relevant information at the time of adjudication. The Clinical Severity Adjudication Committee's decisions will be considered the definitive classification of the case. The role of the Committee and adjudication process will be provided in the committee's charter and more details regarding the impact on the analysis will be provided in the Statistical Analysis Plan (SAP).

8.1.5. Immunogenicity Assessments

Venous blood samples will be collected for assessment of humoral and cellular immune responses. Time points are detailed in the Schedule of Activities (Section 1.3).

If the participant is unable to complete the study without withdrawing consent, immunogenicity samples will be taken at the early exit visit, but only if the early exit visit is at least 10 days after the previous immunology blood draw. See Section 1.3 for further details.

From external sources, the sponsor will obtain serum samples of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used to assess NI of the seropositivity rate induced by a Ad26.COV2.S booster following a primary Pfizer BNT162b2 regimen to the seropositivity post a primary Pfizer BNT162b2 regimen.

Humoral and cellular immunogenicity assays may include, but are not limited to, the assays summarized in Table 4 and Table 5, respectively.

Table 4: Summary of Humoral Immunogenicity Assays

Assay	Purpose
Primary/Secondary endpoints	
SARS-CoV-2 neutralization (VNA)	Analysis of neutralizing antibodies to the live SARS-CoV-2 original strain and leading variant of high consequence or concern*, using a live VNA and/or pseudovirion expressing S protein neutralization assay
Secondary endpoints	
SARS-CoV-2 neutralization (VNA)	Analysis of neutralizing antibodies to additional relevant live SARS-CoV-2 variants of concern and/or pseudovirion expressing S protein from SARS-CoV-2 variants of concern
SARS-CoV-2 binding antibodies (ELISA and/or MSD)	Analysis of antibodies binding to SARS-CoV-2 or individual SARS-CoV-2 proteins (eg, S and RBD proteins from the SARS-CoV-2 original strain, leading variant of high consequence or concern* and/or other relevant variants of concern)
Exploratory endpoints	
SARS-CoV-2 neutralization (neutralization assay)	Analysis of neutralizing antibodies to the vaccine strain (or other strain), as measured by an alternative neutralization assay (different from the VNA used for the primary and secondary endpoint)
Adenovirus neutralization (neutralization assay)	Analysis of neutralizing antibodies to adenovirus
SARS-CoV-2 binding antibodies	Analysis of antibodies binding to SARS-CoV-2 or individual SARS-CoV-2 proteins (eg, S and RBD proteins from the SARS-CoV-2 original strain, leading variant of high consequence or concern* and/or other relevant variants of concern) measured by an alternative binding antibody assay (different from the binding antibody assay used in the secondary endpoints)
Functional and molecular antibody characterization	Analysis of antibody characteristics including Fc-mediated viral clearance, avidity, Fc characteristics, Ig subclass and IgG isotype directed against the original SARS-CoV-2 strain, leading variant of high consequence or concern* and/or other variants of concern
Epitope-specificity characterization	Analysis of site-specificity, epitope mapping
Cytokine profiling	Analysis of cytokines, chemokines, and other proteins of the innate or adaptive immune response in the serum or plasma
Passive transfer	Analysis of immune mediators correlating with protection against experimental SARS-CoV-2 challenge in a suitable animal model

ELISA enzyme linked immunosorbent assay; Ig immunoglobulin; MSD Meso Scale Discovery; RBD receptor binding domain; S spike; SARS CoV 2 severe acute respiratory syndrome coronavirus 2; VNA virus neutralization assay
* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Table 5: Summary of Cellular Immunogenicity Assays

Assay	Purpose
Exploratory endpoints	
Flow cytometry (ICS)	Analysis of T cell responses to SARS-CoV-2 S protein peptides from the original strain, leading variant of high consequence or concern* and/or other variants of concern by ICS including $CD4^+$/$CD8^+$, IFNγ, IL-2, TNFα, IL-4, IL-5, IL-13, and/or other Th1/Th2 markers (if feasible)
ELISpot	IFNγ and IL-4 responses to SARS-CoV-2 S protein peptides by PBMCs (if feasible), based on single or dual ELISpot
Gene expression analysis	Analysis of gene expression by RNA transcript profiling and/or analysis of protein translates, in cells or whole blood stimulated with SARS-CoV-2S protein peptides or in unstimulated cells or whole blood (ex vivo).

ELISpot enzyme linked immunospot (assay); ICS intracellular cytokine staining; IFNγ interferon gamma; IL interleukin; PBMC peripheral blood mononuclear cell; S spike; SARS CoV 2 severe acute respiratory syndrome coronavirus 2; Th: T helper; TNFα tumor necrosis factor alpha
* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

8.2. Exploratory Efficacy Assessments

Identification and molecular confirmation of SARS-CoV-2 infection and symptomatic COVID-19 will be performed throughout the study as described in Section 8.1.2 and adjudicated as described in Section 8.1.4. The ePRO to evaluate vaccine efficacy parameters will be the SIC.

Case Definition for Mild COVID-19

- A SARS-CoV-2 positive RT-PCR or molecular test result from any available respiratory tract sample (eg, nasal swab sample, sputum sample, throat swab sample, saliva sample) or other sample;

  AND at any time during the course of observation[a]:

- One of the following symptoms: fever (≥38.0°C or ≥100.4°F), sore throat, malaise (loss of appetite, generally unwell, fatigue, physical weakness), headache, muscle pain (myalgia), gastrointestinal symptoms, cough, chest congestion, runny nose, wheezing, skin rash, eye irritation or discharge, chills, new or changing olfactory or taste disorders, red or bruised looking feet or toes, or shaking chills or rigors.

A case is considered mild when it meets the above case definition but not the moderate to severe/critical definition.

Case Definition for Moderate COVID-19

- A SARS-CoV-2 positive RT-PCR or molecular test result from any available respiratory tract sample (eg, nasal swab sample, sputum sample, throat swab sample, saliva sample) or other sample **AND at any time during the course of observation:**

Any 1 of the following new or worsening signs or symptoms:

- Respiratory rate ≥20 breaths/minute
- Abnormal saturation of oxygen (SpO_2) but still >93% on room air at sea level*
- Clinical or radiologic evidence of pneumonia
- Radiologic evidence of deep vein thrombosis (DVT)
- Shortness of breath or difficulty breathing

OR

Any 2 of the following new or worsening signs or symptoms:

- Fever (≥38.0°C or ≥100.4°F)
- Heart rate ≥90 beats/minute
- Shaking chills or rigors
- Sore throat
- Cough
- Malaise as evidenced by 1 or more of the following**:
  - Loss of appetite
  - Generally unwell
  - Fatigue
  - Physical weakness
- Headache
- Muscle pain (myalgia)
- Gastrointestinal symptoms (diarrhea, vomiting, nausea, abdominal pain)**
- New or changing olfactory or taste disorders
- Red or bruised looking feet or toes

* SpO_2 criteria will be adjusted according to altitude, per the investigator judgment.

** Having 2 or more elements of a symptom (eg, vomiting and diarrhea or fatigue and loss of appetite) is counted only as 1 symptom for the case definition. To meet the case definition, a participant would need to have at least 2 different symptoms.

Case Definition for Severe/Critical COVID-19

- A SARS-CoV-2 positive RT-PCR or molecular test result from any available respiratory tract sample (eg, nasal swab sample, sputum sample, throat swab sample, saliva sample) or other sample

AND any 1 of the following at any time during the course of observation[a]:

- Clinical signs at rest indicative of severe systemic illness (respiratory rate ≥30 breaths/minute, heart rate ≥125 beats/minute, oxygen saturation (SpO_2) ≤93% on room air at sea level*, or partial pressure of oxygen/fraction of inspired oxygen (PaO_2/FiO_2) <300 mm Hg)

  * SpO_2 criteria will be adjusted according to altitude per the investigator judgment.
- Respiratory failure (defined as needing high-flow oxygen, non-invasive ventilation, mechanical ventilation, or extracorporeal membrane oxygenation [ECMO])
- Evidence of shock (defined as systolic blood pressure <90 mm Hg, diastolic blood pressure <60 mm Hg, or requiring vasopressors)
- Significant acute renal, hepatic, or neurologic dysfunction
- Admission to the ICU
- Death

Case Definition for Asymptomatic COVID-19

Asymptomatic cases of COVID-19, as determined by seropositivity for the SARS-CoV-2 N protein.

8.3. Safety Assessments

Details regarding the IDMC are provided in Committees Structure in Section 10.3.6.

Adverse events will be reported and followed by the investigator as specified in Section 8.4 and Appendix 4.

Any clinically relevant changes occurring during the study must be recorded on the AE section of the eCRF.

[a] Participants will be asked to undertake the COVID-19 procedures until 14 days after symptom onset (COVID-19 Day 15) or until **resolution of the COVID-19 episode**, whichever comes last (see Section 8.1.2). For participants experiencing COVID-19 episodes close to the end of their study participation, COVID episode procedures will be followed up to their EoT visit.

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.

For participants undergoing COVID-19 episodes near, or at the end of the trial participation, the End of Trial (EoT visit) should not be delayed in order to capture full illness episode. 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 (see Section 8.4.1 for further details).

The study will include the following evaluations of safety and reactogenicity according to the time points provided in the Schedule of Activities (Section 1.3).

8.3.1. Physical Examinations

A history-directed physical examination, including height and body weight, will be carried out at screening. To obtain the actual body weight, participants must be weighed lightly clothed. The height should be measured without footwear.

At all other visits, an abbreviated, symptom-directed examination might be performed by the investigator based on any clinically relevant issues or symptoms, and medical history. Symptom-directed physical examination may be repeated if deemed necessary by the investigator.

A targeted physical examination will be performed during a COVID-19 episode by the investigator or designated medically trained clinician (or a trained HCP, if allowed per local regulations).

Physical examinations will be performed by the investigator or designated medically trained clinician. Any clinically relevant abnormalities or changes in severity observed during the review of body systems should be documented in the eCRF.

8.3.2. Vital Signs

Vital signs will be assessed at the timepoints specified in the Schedule of Activities (Section 1.3). On Day 1, vital signs will be measured before vaccination and after vaccination following the 30 minutes post-vaccination observation period.

Body temperature (oral route preferred, or in accordance with the local standard of care), pulse/heart rate, respiratory rate, and blood pressure will be assessed.

Participants will utilize an e-Diary to record body temperature measurements post-vaccination.

All participants with COVID-19 signs and symptoms should measure body temperature daily (oral route preferred, or in accordance with the local standard of care) and record the highest temperature in the last 24 hours each day in the ePRO in the eCOA, for the duration of follow-up of COVID-19 episodes (as defined in Section 8.1.2).

Vital signs will be measured during a COVID-19 episode by a qualified member of the study site. This includes measurement of preferably supine systolic and diastolic blood pressure, heart rate,

respiratory rate, oxygen saturation, and body temperature. It is recommended that vital signs are measured before collection of nasal swabs and blood draws.

Blood pressure measurements will be assessed in a supine position (preferably) with a completely automated device. Manual techniques will be used only if an automated device is not available.

Blood pressure 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 recommended before blood sampling.

8.3.3. Clinical Safety Laboratory Assessments

Blood samples for clinical laboratory assessments (as detailed in Appendix 2) will be collected as described in the Schedules of Activities in Section 1.3.

In the case of a thrombotic event or TTS, every effort should be made to collect local hospital/laboratory test results obtained by the treating physician to allow rapid diagnosis and treatment. This information should be reported through the AESI form (see Appendix 11) electronically per instructions in the eCRF completion guidelines. In addition, every effort should be made to collect blood samples from the participant for a platelet count (local laboratory or substitute for local laboratory) and other applicable testing (central laboratory) (see the Schedule of Activities in Section 1.3.2 and Appendix 2). The Investigator will review the laboratory test results to assist the investigation of the AESI.

See Section 8.4.7 for details on laboratory test details to be reported for an AE of thrombocytopenia.

8.4. Adverse Events, Serious Adverse Events, Adverse Events of Special Interest, and Other Safety Reporting

Timely, accurate, and complete reporting and analysis of safety information, including AEs, SAEs, AESIs and product quality complaints (PQCs), 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.

Adverse events will be reported by the participant (or, when appropriate, by a caregiver, surrogate, or the participant's legally acceptable representative) for the duration of the study.

Further details on AEs, SAEs, AESIs, and PQCs can be found in Appendix 4.

8.4.1. Time Period and Frequency for Collecting Adverse Event, Adverse Event of Special Interest, and Serious Adverse Event Information

All Adverse Events

Adverse events and special reporting situations, whether serious or non-serious, that are related to study procedures or that are related to non-investigational sponsor products will be reported from the time a signed and dated ICF is obtained until the end of the study/early withdrawal.

Clinically relevant medical events not meeting the above criteria and occurring between signing of ICF and moment of vaccination will be collected on the medical history eCRF page as pre-existing conditions.

Solicited AEs, collected through an e-Diary, will be recorded from the time of vaccination until 7 days post-vaccination.

All other unsolicited AEs and special reporting situations, whether serious or non-serious, will be reported from the time of vaccination until 28 days post-vaccination.

All AESIs, SAEs and AEs leading to discontinuation from the study (regardless of the causal relationship) are to be reported from the moment of vaccination until completion of the participant's last study-related procedure, which may include contact for safety follow-up. The sponsor will evaluate any safety information that is spontaneously reported by an investigator beyond the time frame specified in the protocol.

All AEs will be followed until resolution or until clinically stable.

For participants experiencing a COVID-19 episode near the end of study participation: Procedures outlined in Section 1.3.3 should be followed up to to the EoT visit only. For example, if a COVID-19 episode starts -4 days prior to scheduled EoT visit, the Day 1- Day 2 and Day 3- Day 5 procedures should be followed, as much is possible, and then study procedures should stop. Once a participant has had their EoT visit, the outcome cannot be reported in the eCRF, therefore investigators should record the resolution/outcome of the COVID-19 illness in the appropriate source documents.

Adverse Events of Special Interest

TTS is considered to be an AESI. Suspected AESIs (thrombotic events and thrombocytopenia [defined as platelet count below 150,000/µL (Brighton Collaboration 2021)]) will be recorded from the moment of vaccination until the end of the study/early withdrawal. An AESI Adjudication Committee with appropriate expertise will be established to evaluate each suspected AESI and determine whether it is a case of TTS (see Section 8.4.7).

Serious Adverse Events

All SAEs, as well as PQCs, occurring during the study must be reported to the appropriate sponsor contact person by study site personnel within 24 hours of their knowledge of the event.

Serious adverse events, including those spontaneously reported to the investigator must be reported. The sponsor will evaluate any safety information that is spontaneously reported by an investigator beyond the time frame specified in the protocol.

Information regarding SAEs will be transmitted to the sponsor using the SAE Form and Safety Report Form of the eCRF, which must be completed and reviewed 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 transmitted electronically or by facsimile (fax). Telephone reporting should be the exception and the reporter should be asked to complete the appropriate form(s) first.

8.4.2. Method of Detecting Adverse Events, Adverse Events of Special Interest, and Serious Adverse Events

Care will be taken not to introduce bias when detecting AEs, AESIs, 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 used to assess the reactogenicity of the study vaccine and are predefined local (at injection site) and systemic events for which the participant is specifically questioned and which are noted by participants in their e-Diary.

After booster vaccination, participants will remain under observation at the study site for at least 30 minutes for the presence of any acute reactions and solicited events.

In addition, participants will record solicited signs and symptoms in an e-Diary for 7 days post-vaccination. All participants will be provided with an e-Diary and instructions on how to complete the diary (see Overview in Section 8). Electronic diary information will be transferred from the e-Diary source to the sponsor. After review and verbal discussion of the initial e-Diary entries with the participant, the investigator will complete his/her own assessment in the relevant sections of the eCRF/eCOA. Once a solicited sign or symptom from an e-Diary is considered to be of severity Grade 1 or above, it will be recorded as a solicited AE.

1) Solicited Injection Site (Local) Adverse Events

Participants will be asked to note in the e-Diary occurrences of injection site pain/tenderness, erythema and swelling at the study vaccine injection site daily for 7 days post-vaccination (day of vaccination and the subsequent 7 days). The extent (largest diameter) of any erythema and swelling should be measured (using the ruler supplied) and recorded daily. The case definitions for solicited injection site events can be found in the references (Gidudu 2012; Kohl 2007).

2) Solicited Systemic Adverse Events

Participants will be instructed on how to record daily temperature using a thermometer provided for home use. Participants should record the temperature in the e-Diary in the evening of the day of vaccination, and then daily for the next 7 days approximately at the same time each day. If more

than one measurement is made on any given day, the highest temperature of that day will be used in the eCRF.

Fever is defined as endogenous elevation of body temperature ≥38° C, as recorded in at least one measurement (Marcy 2004).

Participants will also be instructed on how to note signs and symptoms in the e-Diary on a daily basis for 7 days post-vaccination (day of vaccination and the subsequent 7 days), for the following events: fatigue, headache, nausea, and myalgia.

Unsolicited Adverse Events

Unsolicited AEs are all AEs for which the participant is not specifically questioned in the participant e-Diary.

For details about AESIs, refer to Section 8.4.7.

8.4.3. Follow-up of Adverse Events, Adverse Events of Special Interest and Serious Adverse Events

The investigator is obligated to perform or arrange for the conduct of supplemental measurements and evaluations as medically indicated to elucidate the nature and causality of the AE, AESI, SAE, or PQC as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.

Adverse events, including pregnancy, will be followed by the investigator as specified in Appendix 4.

8.4.4. Regulatory Reporting Requirements for Serious Adverse Events

The sponsor assumes 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 Institutional Review Board (IRB) that approved the protocol unless otherwise required and documented by the IRB. A SUSAR will be reported to regulatory authorities unblinded. Participating investigators and IRB will receive a blinded SUSAR summary, unless otherwise specified.

8.4.5. Pregnancy

All initial reports of pregnancy in participants or 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 an SAE reporting form. Any participant who becomes pregnant during the study will remain in the study and will continue to undergo all procedures for follow-up and all safety follow-up as outlined in the protocol for all participants.

Follow-up information regarding the outcome of the pregnancy and any postnatal sequelae in the infant will be required.

8.4.6. Disease-Related Events and Disease-Related Outcomes Not Qualifying as Adverse Events or Serious Adverse Events

Serious adverse events caused by molecularly confirmed SARS-CoV-2 infection will be removed at the analysis level from the (S)AE listings and tables and presented separately.

All events that meet the definition of an SAE will be reported as SAEs, regardless of whether they are protocol-specific assessments.

8.4.7. Adverse Events of Special Interest

Adverse events of special interest (AESIs) are significant AEs that are judged to be of special interest because of clinical importance, known class effects, or based on nonclinical signals. Adverse events of special interest will be carefully monitored during the study by the sponsor.

AESIs must be reported to the sponsor within 24 hours of awareness irrespective of seriousness (ie, serious and non-serious AEs) or causality following the procedure described above for SAEs.

Specific requirements for the AESI are described below.

8.4.7.1. Thrombosis with Thrombocytopenia Syndrome

As described in Section 2.3.1, TTS has been observed very rarely following vaccination with Ad26.COV2.S and is considered to be an AESI in this study. TTS is a syndrome characterized by a combination of both a thrombotic event and thrombocytopenia (American Society of Hematology 2021; Brighton Collaboration 2021).

Because this syndrome is rare and not completely understood, all cases of thrombosis and/or thrombocytopenia will be considered a suspected case of TTS until further adjudication can be performed. An AESI Adjudication Committee with appropriate expertise will be established to evaluate each suspected AESI and determine whether it is a case of TTS. The investigator shall be responsible for reporting any suspected AESI of TTS using the SAE form and the form detailed in Appendix 11. A suspected TTS case is defined as:

- Thrombotic events: suspected deep vessel venous or arterial thrombotic events as detailed in Appendix 10
- Thrombocytopenia, defined as platelet count below 150,000/µL (Brighton Collaboration 2021)

Symptoms, signs, or conditions suggestive of a thrombotic events should be recorded and reported as a suspected AESI even if the final or definitive diagnosis has not yet been determined, and alternative diagnoses have not yet been eliminated or shown to be less likely. Follow-up information and final diagnoses, if applicable, should be submitted to the sponsor as soon as they become available.

In the event of thrombocytopenia, study site personnel should report the absolute value for the platelet count and the reference range for the laboratory test used.

For either a thrombotic event or thrombocytopenia, testing for anti-PF4 should be performed at the local laboratory or substitute local laboratory; repeat testing may be requested for confirmation upon sponsor discretion.

Suspected AESIs will require enhanced data collection and evaluation (see Section 1.3.2). Every effort should be made to report as much information as possible about the AESI to the sponsor in a reasonable timeframe.

If an event meets the criteria for an SAE (Section 10.4.1), it should be reported using the same process as for other SAEs.

The form detailed in Appendix 11 is intended as a guide for assessment of the AESIs to facilitate diagnosis and determine treatment options. If the investigator is not the treating physician, every effort should be made to collect the information requested in the form from the treating physician and enter the available information in the eCRF.

8.5. Virology Assessments

Nasal swabs will be used to detect and/or quantify SARS-CoV-2. Exploratory quantification of the SARS-CoV-2 viral load in saliva samples may also be performed.

Nasal swabs collected during a confirmed COVID-19 episode may also be tested at a central laboratory for the presence of other respiratory pathogens using a broad respiratory pathogens panel.

All confirmed COVID-19 episodes will be communicated to the respective participant and to other authorities according to local regulations.

Participants, with stable/well-controlled HIV infection, will be encouraged to have HIV RNA viral load and CD4 cell count assessed at least twice a year and to provide these data for inclusion in the eCRF.

8.6. Biomarkers

During a COVID-19 episode, blood will be collected on COVID-19 Day 3-5 and on COVID-19 Day 29 for evaluation of biomarkers (eg, those associated with severe COVID-19).

8.7. Medical Resource Utilization and Health Economics

In this study, MRU will be collected by interview with the participant and recorded in the eCRF by the investigator and study-site personnel on COVID-19 Day 3-5 and COVID-19 Day 29 (for all participants during a COVID-19 episode; which is defined to be resolved after having 2 consecutive SARS-CoV-2 negative nasal swabs and 2 consecutive days with no COVID-19-related signs or symptoms; see Section 8.1.2) (Appendix 6). Medical resource utilization data will also be collected through the MA-COV form (Appendix 7). This form will be provided to the

participant at the booster vaccination visit and should be completed by the medical care provider or the study site personnel during medical visits for COVID-19 or COVID-19 complications. Protocol-mandated procedures, tests, and encounters are excluded. The data collected will include:

- Number and duration of medical care encounters, including selected procedures (inpatient and outpatient)
- Duration and type of mechanical ventilation and ECMO use
- Duration of hospitalization (total days length of stay, including duration by wards; eg, ICU)
- Number and character of diagnostic and therapeutic tests and procedures
- Outpatient medical encounters and treatments (including physician or emergency room visits, tests and procedures, and medications)

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 immunogenicity and safety data is outlined below. Specific details will be provided in the Statistical Analysis Plan (SAP).

9.1. Statistical Hypotheses

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively by vaccine group.

Hierarchical testing will be applied to the NI hypotheses of neutralizing antibody response to the original strain after boosting vs the original strain after primary vaccination. The NI tests will be performed on the NI analysis set and must be demonstrated on both co-primary endpoints: GMR and seropositivity rates. In addition, an estimated GMR (GMT Day 15 post-booster/GMT post primary regimen) of >0.8 is required to conclude NI.

If the primary hypothesis 1a, in Cohort 1, is met (either at the interim or primary analysis) the primary hypothesis 1b, in Cohort 1 will be tested, accordingly (either at the interim or primary analysis). If the primary hypothesis 1b is met, then the remaining primary hypotheses in Cohort 1 (Ad26.COV2.S primary vaccination) and Cohort 2 (BNT162b2 primary vaccination) will be tested independently (at the primary analysis) and a Bonferroni correction will be used to correct for the Type I error.

The neutralizing antibody response 14 days post Ad26.COV2.S booster vs the neutralizing antibody response 28 days post primary vaccination with Ad26.COV2.S will be used to test the 4 hypotheses related to Cohort 1 (as shown in Figure 3); the neutralizing antibody response 14 days post Ad26.COV2.S booster vs the neutralizing antibody response 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2 will be used to test the 4 hypotheses related to Cohort 2 (as shown in Figure 3):

Cohort 1

1. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.
2. Non-inferiority of neutralizing antibody response to leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, if feasible.
3. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.
4. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, ≥6 months after single-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), compared to the neutralizing antibody response to the original strain induced by single-dose primary vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level.

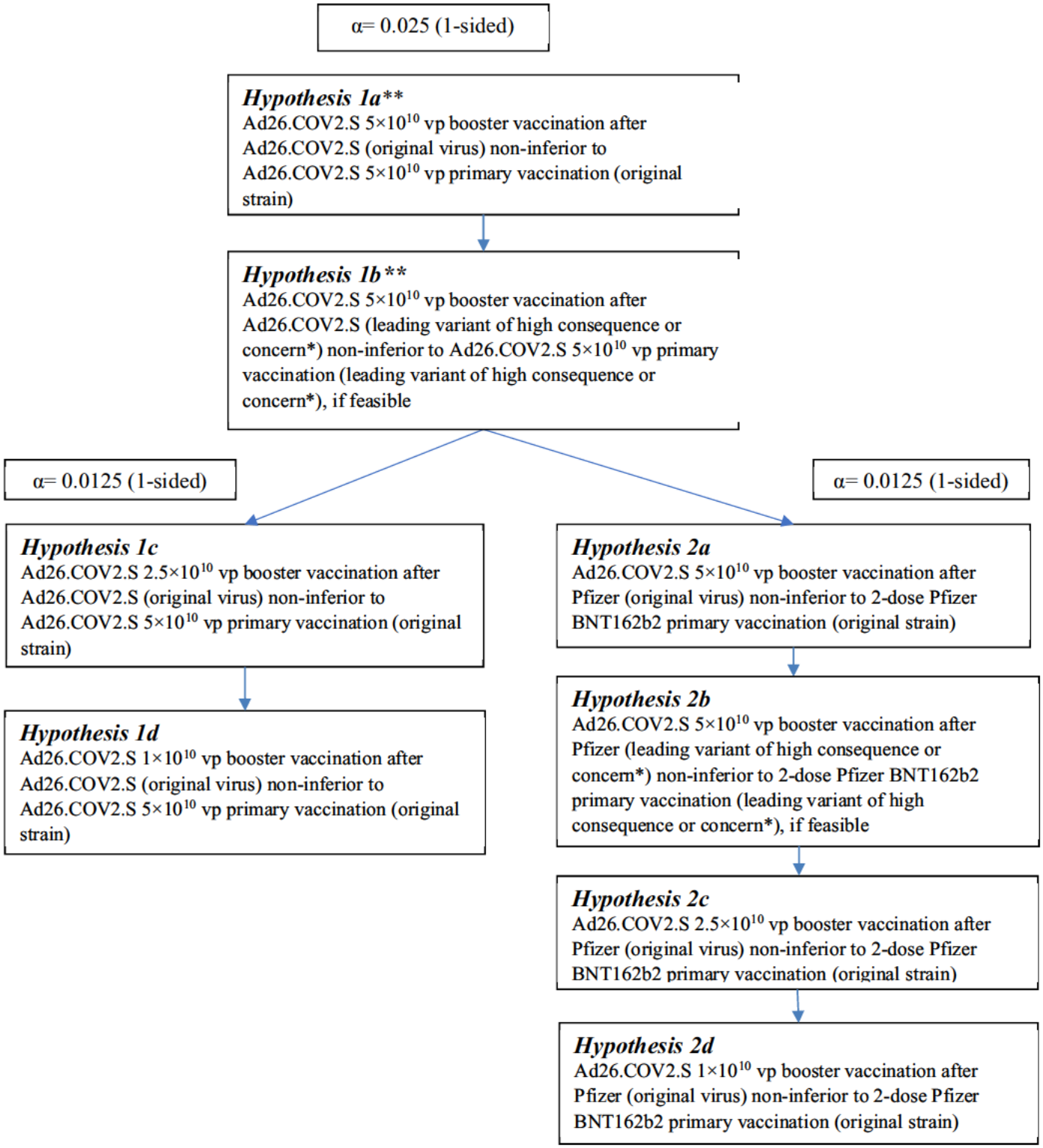
Cohort 2

5. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2.
6. Non-inferiority of neutralizing antibody response to leading variant of high consequence or concern* 14 days after booster vaccination with Ad26.COV2.S at the 5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the leading variant of high consequence or concern* induced by 2-dose primary vaccination with BNT162b2, if feasible.
7. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 2.5×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2.
8. Non-inferiority of neutralizing antibody response to the original strain 14 days after booster vaccination with Ad26.COV2.S at the 1×10^{10} vp dose level, ≥6 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the neutralizing antibody response to the original strain induced by 2-dose primary vaccination with Pfizer BNT162b2.

* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Figure 3 depicts a tree-based schema for testing the non-inferiority hypotheses controlling the FWER at α 0.025 (one-sided) at the interim and primary analyses.

Figure 3: Decision Tree-based Hypothesis Testing



* As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis
** **Note:** at the time of the interim analysis, and if the interim analysis is conducted, only hypotheses 1a and 1b will be tested

In addition to a GMR ratio (GMT Day 15 post-booster/GMT post primary regimen) threshold of >0.8, the following success criteria will be applied to each of the NI tests:

Hypothesis 1a

Success Criterion 1:

The responder rate induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 14 days post booster is NI as compared to the responder rate induced by the Ad26.COV2.S 5×10^{10} vaccine against the original strain 28 days post prime, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 14 days post booster is NI as compared to the GMT induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 28 days post prime, using a NI margin of 1.5-fold.

Note: If primary hypothesis 1a is met at the interim or primary analysis, then primary hypothesis 1b will be tested accordingly (at the interim or primary analysis).

Hypothesis 1b (if feasible)

Success Criterion 1:

The responder rate induced by the Ad26.COV2.S 5×10^{10} vp against the leading strain of high consequence or concern[a] 14 days post booster is NI as compared to the responder rate induced by the Ad26.COV2.S 5×10^{10} vaccine against the leading variant of high consequence or concern* 28 days post prime, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 5×10^{10} vp against the leading strain of high consequence or concern[g] 14 days post booster is NI as compared to the GMT induced by the Ad26.COV2.S 5×10^{10} vp against the leading variant of high consequence or concern* 28 days post prime, using a NI margin of 1.5-fold.

Note: If primary hypothesis 1b is met, then primary hypotheses 1c and 2a will be tested (at the primary analysis).

[a] As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Hypothesis 1c

Success Criterion 1:

The responder rate induced by the Ad26.COV2.S 2.5×10^{10} vp against the original strain 14 days post booster is NI as compared to the responder rate induced by the Ad26.COV2.S 5×10^{10} vaccine against the original strain 28 days post prime, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 2.5×10^{10} vp against the original strain 14 days post booster is NI as compared to the GMT induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 28 days post prime, using a NI margin of 1.5-fold.

Note: If primary hypothesis 1b is met, then primary hypothesis 1c will be tested (at the primary analysis).

Hypothesis 1d

Success Criterion 1:

The responder rate induced by the Ad26.COV2.S 1×10^{10} vp against the original strain 14 days post booster is NI as compared to the responder rate induced by the Ad26.COV2.S 5×10^{10} vaccine against the original strain 28 days post prime, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 1×10^{10} vp against the original strain 14 days post booster is NI as compared to the GMT induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 28 days post prime, using a NI margin of 1.5-fold.

Hypothesis 2a

Success Criterion 1:

The seropositivity rate induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination is NI as compared to the seropositivity rate induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 5×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination is NI as compared to the GMT induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of 1.5-fold.

Note: If primary hypothesis 2a is met, then primary hypothesis 2b will be tested (at the primary analysis).

Hypothesis 2b (if feasible)

Success Criterion 1:

The seropositivity rate induced by the Ad26.COV2.S 5×10^{10} vp against the leading strain of high consequence or concern[a] 14 days post booster is NI as compared to the seropositivity rate induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the leading strain of high consequence or concern[h], using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 5×10^{10} vp against the leading strain of high consequence or concern[g] 14 days post booster is NI as compared to the GMT induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the leading variant of high consequence or concern* 28 days post prime, using a NI margin of 1.5-fold.

Note: If primary hypothesis 2b is met, then primary hypothesis 2c will be tested (at the primary analysis).

Hypothesis 2c

Success Criterion 1:

The seropositivity rate induced by the Ad26.COV2.S 2.5×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination, is NI as compared to the seropositivity rate induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of -10%.

[a] As defined by the Centers for Disease Control and Prevention (CDC Aug 2021c) at the time of the analysis

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 2.5×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination, is NI as compared to the GMT induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of 1.5-fold.

Note: If primary hypothesis 2c is met, then primary hypothesis 2d will be tested (at the primary analysis).

Hypothesis 2d

Success Criterion 1:

The seropositivity rate induced by the Ad26.COV2.S 1×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination, is NI as compared to the seropositivity rate induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of -10%.

Success Criterion 2:

Note: success criterion 2 will only be tested if success criterion 1 was successfully met.

The GMT induced by the Ad26.COV2.S 1×10^{10} vp against the original strain 14 days post booster after 2-dose Pfizer BNT162b2 primary vaccination, is NI as compared to the GMT induced by the 2-dose Pfizer BNT162b2 primary vaccination (external samples) against the original strain, using a NI margin of 1.5-fold.

9.2. Sample Size Determination

The number of participants chosen is to provide sufficient power for the NI comparison stated in Section 9.1. The following assumptions were made in the sample size determinations:

For the non-inferiority comparisons based on seroresponse rate:

- Under the null hypothesis, a 90% responder rate on Day 15 post -booster and a 90% responder rate on Day 29 post primary vaccination
- Non-inferiority margin of -10% for the difference between 14 days post-booster and 28 days post primary vaccination responder rate on Day 29
- Farrington & Manning Likelihood Score Test used in the non-inferiority testing of the difference between the two responder rates

For the non-inferiority comparisons based on GMT:

- Log transformed (log10 scale) VNA data are normally distributed
- A standard deviation (SD) of VNA 0.56 (log10 scale)

- Under the null hypothesis, a GMT ratio (Day 15 post-booster/Day 29 post primary regimen) of 1.0
- Non-inferiority criteria margin log10(2/3) -0.176 for the GMT ratio testing
- No correlation between immune responses 28 days post-dose 1 and 14 days post-booster

In addition, a drop-out rate approximately 10% is assumed.

With the above assumptions, the required sample size to achieve approximately 95% power to demonstrate non-inferiority, at alpha 0.025 (1-sided), of primary hypothesis 1a is N 297 (N 330 adjusted for the 10% dropout). With this sample size, the power to demonstrate non-inferiority of response rate is 98% and the power to demonstrate non-inferiority of the geometric mean ratio (GMR) is 97% resulting in a combined power of approximately 95%.

Similarly, the required sample size to achieve 90% power to demonstrate the non-inferiority, at alpha 0.0125 (1-sided), of the remaining primary hypotheses is N 297 (N 330 adjusted for the 10% dropout). With this sample size, the power to demonstrate non-inferiority of response rate is 95% and the power to demonstrate non-inferiority of the geometric mean ratio (GMR) is 94%, resulting in a combined power of approximately 90%.

When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort 1 Group 1 participants. In case the interim analysis is conducted, an O'Brien-Fleming adjustment will be used whereby the type I error for the non-inferiority test at the interim analysis is 0.0003 (one-sided) and at the final analysis 0.0249. Since an O'Brien-Fleming adjustment is used, the total sample size is not increased for this interim analysis.

The statistical software PASS 15 (version 15.0.5) was used in the sample size calculations. In addition, Addplan version 6.1.1 was used to determine the alpha spending for the interim analysis.

9.3. Analysis Sets

For purposes of analysis, the following populations are defined:

FAS: The full analysis set will include all participants with a documented study vaccine administration (Ad26.COV2.S). Analyses of safety and reactogenicity will be performed on the FAS.

PPI: The per protocol immunogenicity population will include all vaccinated participants for whom post-baseline immunogenicity data are available excluding participants with major protocol deviations expected to impact the immunogenicity outcomes. In addition, samples obtained after natural SARS-CoV-2 infection (if applicable) will be excluded from the analysis.

NI: The NI analysis set will include all PPI participants who are SARS-COV-2 seronegative at baseline (based on the serological test for SARS CoV-2-specific nucleocapsid antibodies [N serology]). The NI hypothesis tests, as well as descriptive analysis of secondary endpoints for other VOCs, will be performed on the NI analysis set.

PPE: The per protocol for efficacy analysis set will include all vaccinated participants who are SARS-CoV-2 seronegative at baseline and who have no major protocol deviations expected to impact the efficacy outcomes.

Pfizer BNT162b2 external samples: serum samples from external sources of approximately 300 individuals collected 2 weeks to 2 months after completion of the 2-dose primary vaccination with Pfizer BNT162b2. These samples will be used for NI assessments for Cohort 2.

9.4. Statistical Analyses

The SAP will be finalized prior to the database lock of the interim analysis, if conducted. If the interim analysis is not conducted, then the SAP will be finalized prior to the database lock of the primary analysis. The SAP will include a more technical and detailed description of the statistical analyses described in this section. This section is a summary of the planned statistical analyses of the most important endpoints including primary and key secondary endpoints.

9.4.1. General Considerations

For safety and immunogenicity analyses, results will be analyzed by vaccine group (using the "as treated principle", ie, by actually received vaccine).

If a participant has a positive SARS-CoV-2 test within 28 days after vaccination, this participant will remain in the study but will not be included in the immunogenicity analysis.

9.4.2. Primary Endpoints

Immunogenicity Endpoints

For NI assessments of primary objectives 1a and 1b (evaluated at the interim or primary analysis), the Ad26.COV2.S booster vaccine at the 5×10^{10} vp dose level against the original strain (objective 1a) and leading variant of concern (objective 1b) as compared to the Ad26.COV2.S vaccine against the original strain (objective 1a) and leading variant of concern (objective 1b) at 28 days post-vaccination, two NI hypothesis tests will be performed:

1. Statistical NI of responder rate, with a margin of -10%: the difference in responder rate between post booster minus post prime will be estimated, with a 100*(1-2*α)% CI. Non-inferiority for responder rate will be demonstrated if the 2-sided 100*(1-2*α)% CI lies entirely above -10%.
2. Statistical NI of GMTs, with a margin of 1.5-fold: the ratio of the GMTs (post booster GMT divided by post prime GMT, i.e; the Geometric Mean Ratio [GMR]) will be estimated, with the corresponding 100*(1-2*α)% CI. Non-inferiority will be demonstrated if the 2-sided 100*(1-2*α)% CI of the estimated GMT ratio lies entirely above 2/3.

3. In addition to the above, an estimated GMR (GMT Day 15 post-booster/GMT Day 29 post primary regimen) of >0.8 is required to conclude NI.

For objectives 1a and 1b, at the interim analysis (if conducted), an O'Brien-Fleming adjustment will be used, α 0.0003 and a 99.4% CI will be calculated. At the primary analysis, α 0.0249 and a 95.02% CI will be calculated, if the interim analysis is conducted. Otherwise, α 0.025 and a 95% CI will be calculated.

Similar non-inferiority criteria will be used for the remaining primary objectives 1c, 1d, 2a, 2b, 2c and 2d (evaluated only at the primary analysis), except that a 97.5% CI will be used for both the responder rate and GMT hypothesis testing.

A hierarchical testing strategy will be applied as depicted in Figure 3.

For primary objectives 1a, 1b, 1c and 1d, formal NI testing will be conducted as a "within-subjects" analysis, in which participants' VNA data are considered matched pairs across the two time points. The NI analysis on responder rate will be based on Agresti-Min (Agresti 2005) method to estimate the difference in proportions and its CI. This method was chosen because of its well-behaved CP properties compared to other methods for the analysis of matched pairs data (Reed 2009). Coverage probability is generally used to evaluate (1 α) CIs where α is the Type I error rate. The NI analysis on the GMT ratio (GMR) will use a paired t-test to estimate the difference in means and its CI on log10 transformed data. The estimated difference and its CI will be back transformed to yield the GMR and its CI.

As a sensitivity analysis, 28 days post-dose 1 VNA data may be pooled across the three study arms and compared to the 14 days post booster VNA data in the study arm of interest. This sensitivity analysis will use appropriate statistical models, such as linear mixed models for the GMR and Generalized Estimating Equations (GEE) models for the difference in responder rates.

For primary objectives 2a, 2b, 2c and 2d relating to Cohort 2, formal NI testing will be conducted by comparing VNA data from external samples, collected 2 weeks to 2 months after completing a 2-dose primary vaccination with Pfizer BNT162b2, compared to the 14 days post booster VNA data in the study arm of interest (groups 4, 5 or 6).

9.4.3. Secondary Endpoint(s)

Safety Endpoints

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively by vaccine group. All safety analyses will be made on the FAS.

When 330 participants are vaccinated, the observation of 0 reactions would be associated with a 95% confidence that the true rate is less than 0.9%. Table 6 provides the probabilities of observing at least one AE at given true AE rates.

Table 6: Probability of Observing at Least One Adverse Event Given a True Adverse Event Incidence

True Adverse Event Incidence	Probability of Observing at Least One Adverse Event	
	N*=330	N*=110
0.1%	28%	10%
0.5%	81%	42%
1%	96%	67%
2.5%	100%	94%
5%	100%	>99%

N: number of participants receiving active vaccine
* N/treatment group: 330 participants in Cohort 1, treatment groups 1 and 2 and Cohort 2, treatment groups 4 and 5; 110 participants in treatment Cohort 1, treatment group 3 and Cohort 2, treatment group 6.

Detailed statistical methodology for analysis of secondary endpoints will be described in the SAP.

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 with onset during the active vaccination phase (ie, AEs occurring after vaccination up to 28 days post-vaccination), SAEs and AESIs will be included in the analysis. (S)AEs caused by molecularly confirmed SARS-CoV-2 infection will be removed at the analysis level from the (S)AE listings and tables and presented separately. For each AE, the percentage of participants who experience at least 1 occurrence of the given event will be summarized by vaccine group.

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

Solicited local (at injection site) and systemic AEs will be summarized descriptively. The number and percentages of participants with at least one solicited local (at injection site) or systemic AE will be presented. The frequencies by vaccine group as well as frequencies according to severity and duration will be described for solicited AEs. Frequencies of unsolicited AEs, separately for all and vaccination-related only, will be presented by System Organ Class and preferred term, while those of solicited AEs will be presented only by preferred term.

Clinical Laboratory Tests

Laboratory data (abnormal or graded, when available) will be listed and/or tabulated by participant and time point.

Vital Signs

Vital signs including temperature, pulse/heart rate, respiratory rate, and blood pressure (systolic and diastolic) will be summarized over time, using descriptive statistics and/or graphically. The percentage of participants with values beyond clinically important limits will be summarized.

Physical Examinations

Physical examination findings will be summarized at baseline. Physical examination abnormal findings will be listed.

Immunogenicity Endpoints

Descriptive statistics (geometric mean and CIs, or median and interquartile range Q1-Q3, as appropriate) will be calculated for continuous immunologic parameters. Several definitions of serological response will be applied as applicable (GMC [S-ELISA], GMT [VNA]). Graphical representations of immunologic parameters will be made as applicable. Frequency tabulations will be calculated for discrete (qualitative) immunologic parameters, as applicable.

In addition, the ratio between neutralizing and binding antibodies as determined by VNA and S-ELISA, respectively, will be calculated together with CIs.

As described in Section 9.1, the powered primary NI endpoints are based on neutralizing antibodies to the original strain or to the leading variant of high consequence or concern (as defined by the Centers for Disease Control and Prevention [CDC Aug 2021c] at the time of the analysis). Additional descriptive analyses will be conducted on other variants, as available. These descriptive analyses will be conducted on the NI set and will include assessments that allow interpretation of the data using NI criteria, but these assessments will not constitute formal hypothesis testing.

For Cohort 1, these assessments will include, but are not limited to:

- Geometric Mean Titer Ratio (with 95% CI) from 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination to 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), using a paired t-test
- Difference in % responders (with 95% CI) between 28 days after Ad26.COV2.S (5×10^{10} vp dose level) single-dose primary vaccination and 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 1-dose primary vaccination with Ad26.COV2.S (5×10^{10} vp dose level), using the Agresti-Min method (Agresti 2005)

Note: as these analyses refer to two time points, only study participants with data at both time points will contribute to these analyses. Within-subjects analysis methods (paired t-test and Agresti-Min) will be used.

For Cohort 2, these assessments will include, but are not limited to:

- Geometric Mean Titer Ratio (with 95% CI) of the neutralizing antibody responses 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 to the neutralizing antibody responses in serum samples of approximately 300 individuals, collected 2 weeks to 2 months after completing 2-dose primary vaccination with Pfizer BNT162b2 (also referred to as Pfizer BNT162b2 external samples), using a t-test
- Difference in % seropositive participants (with 95% CI) between 14 days after Ad26.COV2.S booster vaccination (5×10^{10} vp dose level) after completing 2-dose primary vaccination with Pfizer BNT162b2 and the Pfizer BNT162b2 external samples, using the Farrington-Manning test

Note: in contrast to Cohort 1, the use of external samples in Cohort 2 implies that between subjects analysis methods are to be used in this cohort (t-test and Farrington-Manning test).

The immunogenicity analyses will be performed on the PPI population and the FAS; selected immunogenicity analyses will be performed on the NI analysis set.

9.4.4. Exploratory Endpoint(s)

If feasible, rVE of booster vaccination with Ad26.COV2.S may be estimated at the time of the final analysis or any interim analysis conducted after the primary analysis, if the required data are available, and a sufficient number of cases are recorded. The following rVE estimates may be calculated:

- Within Cohort 1 and Cohort 2 separately: the rVE of Ad26.COV2.S 5×10^{10} vp compared to Ad26.COV2.S 1×10^{10} vp and the rVE of Ad26.COV2.S 2.5×10^{10} vp compared to Ad26.COV2.S 1×10^{10} vp
- The rVE of Ad26.COV2.S 5×10^{10} vp in Cohort 2 compared to Cohort 1
- The rVE of Ad26.COV2.S 2.5×10^{10} vp in Cohort 2 compared to Cohort 1
- The rVE Ad26.COV2.S 1×10^{10} vp in Cohort 2 compared to Cohort 1

The endpoints will be:

- COVID-19 cases meeting the moderate case definitions
- COVID-19 cases meeting the moderate to severe/critical case definitions
- COVID-19 cases meeting the severe/critical case definition
- COVID-19 cases meeting the US FDA Harmonized COVID-19 case definition
- Cases meeting the asymptomatic/undetected case definition
- Case counting will start 14 days post vaccination in COV2008.

The analysis will be based on the Cox proportional hazards model, using the calendar-based time scale. If feasible, the model may stratify for site or a higher-level geographical region, to account for potential differences in geographical exposure. In addition, the model(s) may add other relevant stratification factors such as age group and/or presence/absence of comorbidities.

Furthermore, additional descriptive analyses may be provided. These analyses may include but are not limited to:

- The number of symptoms reported by homologous/heterologous vaccination regimen and by dose level
- The number and percentage of severe/critically ill cases by homologous/heterologous vaccination regimen and by dose level

Additional details will be provided in a supplemental SAP.

9.4.5. Other Analyses

If feasible, further exploratory analyses may be conducted to investigate the effect of the immunogenicity responses on the probability of experiencing a COVID-19 event (eg, RNA-seq

responses, immunological correlates of protection/risk against SARS-CoV-2 infection if feasible). These analyses may utilize logistic or probit regression models, in which statistical control for potential confounders such as age and sex may be included. Additionally, linear regression or other suitable models may be explored to investigate the effect of the dose level on the mounted immune responses, while statistically controlling for potential confounders.

Statistical analysis of biomarker responses will be detailed in a separate SAP.

9.5. Planned Analysis

Interim Analysis

When at least 330 participants (see Section 4.1) from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit, and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis may be conducted whereby, if conducted, the formal non-inferiority testing of Cohort 1 Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort 1 Group 1 participants.

Scope of the Interim Analysis:

If conducted, the interim analysis will consist of a statistical immunogenicity analysis of Cohort 1 Group 1 participants only. Hypotheses 1a and 1b will be tested, in the hierarchical manner as outlined in Figure 3: Hypothesis 1b will only be tested if all success criteria of Hypothesis 1a are met. The other hypotheses (1c, 1d, 2a, 2b, 2c and 2d) will only be tested at the primary analysis.

In addition, limited safety/reactogenicity statistical outputs will be generated in a blinded manner, for all available participants (Cohort 1 and Cohort 2, all groups). The same outputs will be generated in an unblinded manner by the independent Statistical Support Group. The unblinded outputs may also be used to respond to questions or requests from Health Authorities. In this case, appropriate channels of communication will be used to keep the Sponsor blinded.

Alpha Spending:

If the interim analysis is conducted, an O'Brien-Fleming adjustment will be used whereby the type I error for the NI test at the interim analysis will be 0.0003 (one-sided). This alpha is calculated based on an estimated interim analysis sample size of 110 Cohort 1 Group 1 participants (~33% of the total sample size). If immunogenicity data is available for more than 110 Cohort 1 Group 1 participants at the time of the interim analysis, then the nominal alpha level of the interim analysis will be kept at 0.0003, but the alpha level at the primary analysis will be recalculated. The recalculation will be based on the O'Brien-Fleming adjustment and will take into consideration the alpha spent at the time of the interim analysis and the actual information fraction that was available at the time.

Procedures to maintain the study blind:

If performed, this interim analysis will be conducted by an independent Statistical Support Group. Only group-level unblinded immunogenicity statistical outputs and blinded safety/reactogenicity statistical outputs will be available to the Sponsor.

In order to keep the blind at the laboratory, all available Cohort 1 immunogenicity samples, from all groups, will be shipped and analyzed, if operationally feasible. In case a selection of samples is needed for operational reasons, the selection will be made in advance by the independent Statistical Support Group and this selection will include, in addition to the Cohort1 Group 1 samples, also at least 20 samples of Cohort 1 Group 2 and at least 20 samples of Cohort 1 Group 3.

Primary and Final Analyses

The primary and final analyses of Cohort 1 may be performed before the analyses of Cohort 2.

The primary analysis of safety and immunogenicity in Cohort 1 or Cohort 2 will be performed when all evaluable participants in the respective cohort have completed the visit that takes place 28 days after study vaccination, or discontinued earlier. The analysis will include immunogenicity data (VNA, N-serology, and S-ELISA and/or MSD [if available at the time of analysis]) for all evaluable participants through Day 15, and all available safety data. The sponsor will be unblinded at the time of this primary analysis, with the exception of specific sponsor personnel (see below). If the primary analysis of Cohort 1 is performed before the primary analysis of Cohort 2, then the sponsor will be unblinded only for Cohort 1 at the time of the Cohort 1 primary analysis, and unblinded for only Cohort 2 at the time of the primary analysis of Cohort 2.

For the primary analysis, and for any analyses after the primary analysis on the same cohort(s), unblinded data at the participant level will be available to sponsor personnel including statistical programming, statistics, clinical and clinical immunology personnel involved in the analysis, and the sponsor committee involved in making future decisions for the program. Sponsor personnel directly involved in data collection, data management, and safety monitoring () will not have access to unblinded data at the participant level until the study end. Sites and study participants will not have access to unblinded data at the participant level until the end of study (except for events such as emergency unblinding). Group level data may be shared with investigators or other blinded clinical staff, as needed, but every effort will be made to preserve the blinding to the individual participant allocation until the end of study.

The final analysis in Cohort 1 or Cohort 2 will be performed when all included participants in the respective cohort have completed their last visit or discontinued earlier.

After the primary analysis, additional interim analyses may be performed by the sponsor as required.

There is no formal assessment of efficacy in this study. However, data on molecularly confirmed SARS-CoV-2 infections, SARS-CoV-2 asymptomatic infection (by measurement of nucleocapsid binding antibodies), moderate, moderate to severe/critical COVID-19 disease, medical

intervention/hospitalization and COVID-19-like signs and symptoms will be reported after the primary analysis, and rVE may be estimated if feasible from the available data (see Section 9.4.4).

The SAP, and supplemental SAP, will describe the planned analyses in greater detail.

10. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1. Appendix 1: Abbreviations and Definitions

Ad26	adenovirus type 26
ACE2	angiotensin-converting enzyme 2
AdVAC®	adenoviral vaccine (vector platform)
AE	adverse event
AESI	adverse event of special interest
ALT	alanine aminotransferase
ARDS	acute respiratory distress syndrome
ART	anti-retroviral treatment
AST	aspartate aminotransferase
BIDMC	Beth Israel Deaconess Medical Center
BMI	body mass index
BNT162b2	Pfizer mRNA- based SARS-CoV-2 vaccine
BUN	Blood Urea Nitrogen
CDC	Centers for Disease Control and Prevention
CI	confidence interval
COPD	chronic obstructive pulmonary disease
COVID-19	coronavirus disease-2019
CP	coverage probability
CPK	creatine phosphokinase
CS	cytokine storm
CSAC	Clinical Severity Adjudication Committee
CVST	cerebral venous sinus thrombosis
DNA	deoxyribonucleic acid
ECMO	extracorporeal membrane oxygenation
DVT	deep vein thrombosis
eCOA	electronic clinical outcome assessment
eCRF	electronic case report form
eDC	electronic data capture
EEA	European Economic Area
EF-PPND	embryo-fetal and pre- and postnatal development
EoT	End of trial
ePRO	electronic patient-reported outcomes
ELISA	enzyme-linked immunosorbent assay
ELISpot	enzyme-linked immunospot
EMA	European Medicines Agency
EU	European Union
FAS	full analysis set
Fc	crystallizable fragment
FDA	Food and Drug Administration
FI	formalin-inactivated
FOIA	Freedom of Information Act
FWER	family wise error rate
GCP	good clinical practice
GMR	geometric mean ratio
GLP	good laboratory practice
GMC	geometric mean concentration
GMT	geometric mean titer
HA	Health Authority
HCP	health care professional
HIT	heparin-induced thrombocytopenia
HIV	human immunodeficiency virus
HPV	human papillomavirus
IB	investigator's brochure
ICF	informed consent form

ICH	International Conference on Harmonisation
ICS	intracellular cytokine staining
ICMJE	International Committee of Medical Journal Editors
ICU	intensive care unit
IDMC	Independent Data Monitoring Committee
IFN-γ	interferon gamma
Ig	immunoglobulin
IL	interleukin
IM	intramuscular
IND	Investigational New Drug
IPPI	Investigational Product Preparation Instructions
IRB	Institutional Review Board
IWRS	interactive web response system
MA-COV	medically-attended COVID-19
MAH	Marketing Authorization Holders
MedDRA	Medical Dictionary for Regulatory Activities
MERS(-CoV)	Middle East respiratory syndrome (coronavirus)
MSD	Meso Scale Discovery
MIS	multisystem inflammatory syndrome
MRU	medical resource utilization
N	nucleocapsid
NHP	non-human primate
NI	non-inferiority
PCR	polymerase chain reaction
PE	pulmonary embolism
PI	principal investigator
PPE	per protocol efficacy
PPI	per protocol immunogenicity
PQC	product quality complaint
PT	prothombin time
rVE	relative vaccine efficacy
VNA	virius neutralization assay
PRO	patient reported outcomes
RBD	receptor-binding domain
RNA	ribonucleic acid
RSV	respiratory syncytial virus
RT-PCR	real-time reverse-transcriptase polymerase chain reaction
S	spike
SAE	serious adverse event
SAP	statistical analysis plan
SARS	severe acute respiratory syndrome
SARS-CoV(-2)	severe acute respiratory syndrome coronavirus(-2)
SD	standard deviation
SIC	Symptoms of Infection with Coronavirus-19
SIPPM	site investigational product and procedures manual
SRP	study responsible physician
SRS	study responsible scientist
SUSAR	suspected unexpected serious adverse reaction
Th	T-helper
TNF-α	tumor necrosis factor alpha
TTS	thrombosis with thrombocytopenia syndrome
ULN	upper limit of the normal range
UK	United Kingdom
US	United States
VAED	vaccine-associated enhanced disease
VAERD	vaccine-associated enhanced respiratory disease
VNA	virus neutralization assay
vp	virus particle

VTE	venous thromboembolism
WBC	white blood cell
WHO	World Health Organization

Definitions of Terms

COVID-19	COVID-19 is the disease caused by the virus SARS-CoV-2. COVID-19 refers to SARS-CoV-2 infection with symptoms, and can range from mild to severe disease, the latter including pneumonia, severe acute respiratory syndrome, multi-organ failure, and death (US FDA 2020b; US FDA 2021a).
e-COA	An umbrella term encompassing different types of outcomes assessments, in particular, the COVID-19 signs and symptoms surveillance question, the ePRO and the e-Diary.
ePRO	The electronic technology used to collect the patient-reported outcome data. PROs are reports that come directly from the participant without interpretation by clinician or anyone else. This includes the SIC questionnaire (Symptoms of Infection with Coronavirus-19) and the recording of pulse oximetry results.
e-Diary	The electronic technology used to record solicited signs and symptoms by the participants.
Study Name Convention	In this document, studies are referred to using the short study name (preceding letters and final digits of the study identifier) only (eg, COV1001).
Electronic source system	Contains data traditionally maintained in a hospital or clinic record to document medical care or data recorded in a CRF as determined by the protocol. Data in this system may be considered source documentation.

10.2. Appendix 2: Clinical Laboratory Tests

The following tests will be performed according to the Section 1.3.1, Section 1.3.2 and Section 1.3.3:

Protocol-Required Safety Laboratory Assessments

Laboratory Assessments	Parameters	Timepoints
Testing done centrally and/or locally	Whole blood sample for platelet count which at some sites may be part of a complete blood count with differential	Pre-booster vaccination and as part of a suspected AESI investigation if applicable
Testing done centrally	Serum/plasma samples for coagulation-related assays such as but not limited to: • Activated partial thromboplastin time • Prothrombin time • International normalized ratio • Fibrinogen • D-dimer • Lupus anticoagulant • Anti-cardiolipin antibody • Beta-2 glycoprotein· • Heparin-induced thrombocytopenia (HIT)/platelet factor (PF) 4 antibody, IgG·(HIT assay) • Platelet activation assay (if HIT/PF4 is positive) • Homocysteine • ADAMTS13 Activity and Inhibitor Profile	Based on the clinical evaluation of the suspected AESI (eg, whether thrombocytopenia is observed in conjunction with a thrombotic event), all or some of these tests may be conducted on the stored pre vaccination sample (retrospective test) and on the samples obtained as part of the AESI investigation. Similar samples from appropriate controls (from vaccinated participants who did not experience an AESI) within the study may be used as part of investigation of AESIs.
Testing done centrally	Serological test for SARS-CoV-2-specific antibodies (based on N serology)	At baseline, Day 15 (all), Day 29 (Subset 2), Day 71 (Subset 3), Day 120 (no subset), Day 181 (all), Day 361 (all)
Testing done either locally or centrally	Nasal swab for virology testing (viral load testing). Negative baseline test is NOT required for enrollment and randomization.	• At baseline (nasal swab collected by qualified study staff and batch tested centrally) • On COVID-19 Day 1-2 (nasal swab collected by the participant at home) • On COVID-19 Day 3-5 (nasal swab collected by qualified study staff) • Once every 2 days following COVID-19 Day 3-5, until closure of the COVID-19 procedures (nasal sample collected by the participant at home)

Laboratory Assessments	Parameters	Timepoints
Testing done centrally	Saliva samples for virology testing (molecular confirmation of SARS-CoV-2 infection and viral load testing), if feasible	• On COVID-19 Day 3-5 (saliva sample collected by the participant at the study site or at home) • Once every 2 days following COVID-19 Day 3-5, until closure of the COVID-19 procedures (saliva sample collected by the participant at home)

10.3. Appendix 3: Regulatory, Ethical, and Study Oversight Considerations

10.3.1. Regulatory and Ethical Considerations

Investigator Responsibilities

The investigator is responsible for ensuring that the study is performed in accordance with the protocol, current International Council for Harmonisation (ICH) guidelines on Good Clinical Practice (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 (LTMs), Clinical Trial Managers (CTMs), and/or Contract Research Organizations (CROs) who will ensure that the PCC explanations are followed by the investigators.

The PCC Document may be shared by the sites with Institutional Review Boards (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 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 IRB and relevant competent authority. Documentation of amendment approval by the investigator and IRB must be provided to the sponsor. When the change(s) involve only logistic or administrative aspects of the study, the IRB (where required) only needs to be notified.

In situations where a departure from the protocol is unavoidable during the study, 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 electronic case report form (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 principal investigator (PI)
- A copy of the dated and signed (or sealed, where appropriate per local regulations), written 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 IRB, including a current list of the 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 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 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 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)
- IB (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 IRB)
- Information regarding funding, name of the sponsor, institutional affiliations, other potential conflicts of interest, and incentives for participants
- Any other documents that the IRB requests to fulfill its obligation

This study will be undertaken only after the 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 IRB and the documents being approved.

During the study the investigator (or sponsor where required) will send the following documents and updates to the 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 IB and amendments/addenda
- Summaries of the status of the study at intervals stipulated in guidelines of the 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 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 IRB for review and approval before implementation of the change(s).

At least once a year, the 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 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 4.2.1.

Other Ethical Considerations

For study-specific ethical design considerations, refer to Section 4.2.1.

10.3.2. Financial Disclosure

Investigators and sub investigators 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 study and for 1 year after completion of the study.

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

10.3.3. 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 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 long-term follow-up 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.

Participants who are rescreened are required to sign a new ICF.

If the participant is unable to read or write, an impartial witness should be present 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 of the participant is obtained.

10.3.4. 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, 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.

10.3.5. 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 vaccination with Ad26-based vaccines, including Ad26.COV2.S, to understand SARS-CoV-2 infection, to understand differential intervention responders, and to develop tests/assays related to Ad26-based vaccines, includingAd26.COV2.S and SARS-CoV-2 infection. 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).

10.3.6. Committees Structure

Independent Data Monitoring Committee

An IDMC consisting of members who are not directly involved in the study conduct, data management, or statistical analysis, has been established. After the primary analysis, The IDMC will review data on an ad hoc basis upon request of the sponsor to ensure the continuing safety of the participants enrolled in this study. The IDMC will review data as indicated in Section 4.1. When appropriate, the conclusions of the IDMC will be communicated to the investigators, the IRB, and the national regulatory authorities.

Ad hoc review may be performed further to the occurrence of any AE/AESI/SAE leading to a study pausing situation as outlined in Section 6.8, or at request of the sponsor's medical monitor or designee. The PI(s) and SRP/S will inform the IDMC of any AE of concern.

Upon ad hoc review, the IDMC will review blinded data first but is entitledto request submission of unblinded data if deemed necessary.

The composition of the IDMC, its responsibilities, authorities and procedures are documented in the IDMC Charter.AESI Adjudication Committee

An AESI Adjudication Committee with appropriate expertise will be established to evaluate each suspected AESI and determine whether it is a case of TTS (see Section 8.4.7). A Charter will be developed to describe the roles and responsibilities of the Committee.

Clinical Severity Adjudication Committee

The Clinical Severity Adjudication Committee will review all suspected cases of COVID-19, except for cases already adjudicated as severe, as well as those requiring medical intervention (such as a composite endpoint of hospitalization, ICU admission, mechanical ventilation, and ECMO, linked to objective measures such as decreased oxygenation, X-ray or CT findings), including onset of cases, taking into account all available relevant information at the time of adjudication. More details will be provided in the charter of the Clinical Severity Adjudication Committee. Re-adjudication will occur if new information becomes available. The last adjudication for a given case will determine the status of the case for analysis. The Clinical Severity Adjudication Committee's assessment will be considered the definitive classification of the case.

10.3.7. Publication Policy/Dissemination of Clinical Study Data

All information, including but not limited to information regarding Ad26.COV2.S 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 Ad26.COV2.S, 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.

10.3.8. 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 may review the 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.

10.3.9. Case Report Form Completion

Case report forms are prepared and provided by the sponsor for each participant in electronic format. All data relating to the study must be recorded in the 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 eCRF, if applicable. Study-specific data will be transmitted in a secure manner to the sponsor.

Worksheets may be used for the capture of some data to facilitate completion of the eCRF. Any such worksheets will become part of the participant's source documents. Data must be entered into the 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.

All participative measurements (eg, questionnaires) will be completed by the same individual who made the initial baseline determinations whenever possible.

If necessary, queries will be generated in the electronic data capture (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.

10.3.10. 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; intervention receipt/dispensing/return records; study vaccine administration information; and date of study completion and reason for early discontinuation of study vaccine or withdrawal from the study, if applicable.

The author of an entry in the source documents should be identifiable. Given that PROs are reports of a patient’s health condition that come directly from the patient, without interpretation by a clinician or anyone else, the responses to ePRO measures entered by study participants into source records cannot be overridden by site staff or investigators.

Participant- and investigator-completed scales and assessments designated by the sponsor (ie, SIC) will be recorded directly into an eDevice and will be considered source data. The participant’s e-Diary used to collect information regarding solicited signs and symptoms after vaccination will

be considered source data. The documentation of the positive RT-PCR result that serves as a trigger to start procedures for COVID-19 follow-up, will be considered source data.

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 other equivalent document).

An eSource system may be utilized, which contains data traditionally maintained in a hospital or clinic record to document medical care (eg, electronic source documents) as well as the clinical study-specific data fields as determined by the protocol. This data is electronically extracted for use by the sponsor. If eSource is utilized, references made to the eCRF in the protocol include the eSource system but information collected through eSource may not be limited to that found in the eCRF.

10.3.11. Monitoring

The sponsor will use a combination of monitoring techniques (central, remote, and/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 may 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.

Central monitoring will take place for data identified by the sponsor as requiring central review.

10.3.12. 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. Remote auditing techniques may also be utilized, if necessary. 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.

10.3.13. 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.

10.3.14. 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/Site 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 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

10.4. Appendix 4: Adverse Events, Serious Adverse Events, Adverse Events of Special Interest, Product Quality Complaints, and Other Safety Reporting: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

10.4.1. Adverse Event Definitions and Classifications

Adverse Event

An AE is any untoward medical occurrence in a clinical study participant administered a pharmaceutical (investigational or non-investigational) product. An AE does not necessarily have a causal relationship with the intervention. 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.

Any respiratory tract infection that is not due to SARS-CoV-2 infection will be reported as an AE if it occurs between the time of any vaccination through the following 28 days. Any respiratory tract infection recorded as an AE in the eCRF will be excluded from any AE analysis if the molecular test is subsequently found to be positive for SARS-CoV-2. Respiratory tract infections arising from SARS-CoV-2 infection will not be reported as (S)AEs in the Clinical Study Report but will be tabulated separately. In general, any (S)AEs caused by molecularly confirmed SARS-CoV-2 infection will be removed at the analysis level from the (S)AE listings and tables and presented separately.

Note: The sponsor collects AEs starting with the signing of the ICF (refer to All Adverse Events under Section 8.4.1).

Serious Adverse Event

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 even if it is a component of the study endpoint (eg, all-cause mortality).

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 Ad26.COV2.S, the expectedness of an AE will be determined by whether or not it is listed in the IB.

10.4.2. 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.

10.4.3. Severity Criteria

All AEs and laboratory data will be coded for severity using a modified version of the FDA grading table, based on version of September 2007 (FDA 2007), included in Appendix 8.

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 severity of solicited signs and symptoms will be graded in the e-Diary by the participant based on the severity assessment provided in the e-Diary and then verified by the investigator using the toxicity grading scale in Appendix 8. (Note: severity of the measured events will be derived from the diameter [for erythema and swelling] and the temperature measurements [for fever]).

10.4.4. 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:

- Overdose of a sponsor study vaccine
- 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)
- Thrombotic and bleeding events associated with low platelets require immediate reporting the Marketing authorization holders (MAH).

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

10.4.5. 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 24hour 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 the participant's discontinuation from 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)

Any event requiring hospitalization (or prolongation of hospitalization) that occurs during participation in the 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 or not the event is expected or associated with the study vaccine, is considered an SAE.

Information regarding SAEs will be transmitted to the sponsor using an SAE reporting form and safety report form of the eCRF, which must be completed and reviewed by a physician from the study site, and transmitted in a secure manner to the sponsor within 24 hours. The initial and follow-up reports of an SAE should be transmitted in a secure manner electronically or by facsimile (fax). Telephone reporting should be the exception and the reporter should be asked to complete the appropriate form(s) first.

Adverse Events of Special Interest

AESIs will be carefully monitored during the study by the sponsor. Suspected AESIs must be reported to the sponsor within 24 hours of awareness irrespective of seriousness (ie, serious and non-serious AEs) or causality assessment, following the procedure described above for SAEs and will require enhanced data collection.

10.4.6. Product Quality Complaint Handling

Definition

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, reliability, or performance of a distributed product, including its labeling, drug delivery system, or package integrity. A PQC may have an impact on the safety and efficacy of the product. In addition, it includes any technical complaints, defined as any complaint that indicates a potential quality issue during manufacturing, packaging, release testing, stability monitoring, dose preparation, storage or distribution of the product or the drug delivery system.

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.

A sample of the suspected product should be maintained under the correct storage conditions until a shipment request is received from the sponsor.

10.4.7. Contacting Sponsor Regarding Safety, Including Product Quality

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

10.5. Appendix 5 Symptoms of Infection with Coronavirus-19 (SIC)

The following questions ask about symptoms people with coronavirus-19 infection may experience. Answer each question carefully by choosing 'yes' if you have experienced the symptom or 'no' if you have not experienced the symptom in the last 24 hours. If you choose 'yes,' select the rating that best matches your experience.

In the last 24 hours, have you experienced…	**Please rate the severity of each symptom you experienced.**
Feeling generally unwell (run down) ☐ **Yes** ☐ **No** If yes, →	How severe was your **feeling (generally unwell or run down)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Fatigue (tiredness) ☐ **Yes** ☐ **No** If yes, →	How severe was your **fatigue (tiredness)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Physical weakness ☐ **Yes** ☐ **No** If yes, →	How severe was your feeling of **physical weakness** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Cough ☐ **Yes** ☐ **No** If yes, →	How severe was your **cough** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Shortness of breath (difficulty breathing) ☐ **Yes** ☐ **No** If yes, →	How severe was your **shortness of breath (difficulty breathing)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Sore throat ☐ **Yes** ☐ **No** If yes, →	How severe was your **sore throat** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Nasal congestion (stuffy nose) ☐ **Yes** ☐ **No** If yes, →	How severe was your **nasal congestion (stuffy nose)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Wheezing (whistling sound while breathing) ☐ **Yes** ☐ **No** If yes, →	How severe was your **wheezing (whistling sound while breathing)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible

In the last 24 hours, have you experienced…	Please rate the severity of each symptom you experienced.
Runny nose ☐ **Yes** ☐ **No** If yes, →	How severe was your **runny nose** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Sneezing ☐ **Yes** ☐ **No** If yes, →	How severe was your **sneezing** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Chest congestion (mucus in chest) ☐ **Yes** ☐ **No** If yes, →	How severe was your **chest congestion (mucus in chest)** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Chest pain/ pressure/tightness ☐ **Yes** ☐ **No** If yes, →	How severe was your **chest pain/pressure/tightness** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Muscle aches/pains ☐ **Yes** ☐ **No** If yes, →	How severe were your **muscle aches or pains** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Joint aches/pains ☐ **Yes** ☐ **No** If yes, →	How severe were the **aches or pains in your joints** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Headache ☐ **Yes** ☐ **No** If yes, →	How severe was your **headache** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Feeling faint ☐ **Yes** ☐ **No** If yes, →	How severe was your **feeling of faintness** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Problems thinking clearly/brain fog ☐ **Yes** ☐ **No** If yes, →	How severe were your **problems thinking clearly/brain fog** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible

In the last 24 hours, have you experienced…	Please rate the severity of each symptom you experienced.
Chills ☐ **Yes** ☐ **No** If yes, →	How severe were your **chills** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Skin rash ☐ **Yes** ☐ **No** If yes, →	How severe was your **skin rash** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Eye irritation/discharge ☐ **Yes** ☐ **No** If yes, →	How severe was your **eye irritation/discharge** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Diarrhea ☐ **Yes** ☐ **No** If yes, →	How severe was your **diarrhea** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Vomiting ☐ **Yes** ☐ **No** If yes, →	How severe was your **vomiting** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Nausea ☐ **Yes** ☐ **No** If yes, →	How severe was your **nausea** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Abdominal/ stomach pain ☐ **Yes** ☐ **No** If yes, →	How severe was your **abdominal/stomach pain** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible
Loss of appetite ☐ **Yes** ☐ **No** If yes, →	How severe was your **loss of appetite** in the last 24 hours? ☐ 0 None ☐ 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 6 ☐ 7 ☐ 8 ☐ 9 ☐ 10 Worst possible

What was your **highest temperature** in the last 24 hours? ____ °C/°F

What method did you use to take your temperature?

□ **oral** □ **armpit** □ **ear** □ **forehead** □ **rectal**

In the last 24 hours, have you experienced…

Uncontrollable body shaking/shivering*
□ **Yes** □ **No**

Decreased sense of smell*
□ **Yes** □ **No**

Decreased sense of taste*
□ **Yes** □ **No**

Red or bruised looking feet or toes*
□ **Yes** □ **No**

*Please rate the severity of your symptoms in the last 24 hours?

- □ No Symptoms
- □ Mild
- □ Moderate
- □ Severe

10.6. Appendix 6: Medical Resource Utilization (MRU) Questionnaire

Version for Confirmed COVID-19 Cases

Participant ID:

Date (dd-mmm-yyyy):

1. Medical consultations

Since onset of the confirmed COVID-19 episode, how many times have you had medical consultations?

	No	Yes	Type of contact (personal consultation/ telemedicine)	If yes, specify the number of visits	Specify number of visits related to COVID-19 or its complications	Indicate a reason for each visit
General Practitioner						
Internal Medicine/Medical Outpatient Department						
Other Specialist (Please specify):						
Other (eg Physiotherapy, Pharmacist for a consultation Please specify:)						

2. Professional home care

Please indicate the need for professional care at home since onset of the confirmed COVID-19 episode

	No	Yes	Type of contact (personal consultation/ telemedicine)	If yes, specify the number of visits	Specify number of visits related to COVID-19 or its complications	Indicate a reason for each type of professional care at home
General Practitioner						
Nurse/ Nurse practitioner						
Internal Medicine/Medical Outpatient Department						
Specialist (Please specify):						
Other (eg Physiotherapy, Pharmacist Please specify:)						
Supplemental oxygen						

3. Hospital Services

Since onset of the confirmed COVID-19 episode, did you visit the hospital?

Yes: ____
No: ____

	No	Yes	If yes, specify number of visits/admissions	Specify number of visits/admissions related to COVID-19 or its complications	Specify the length of each stay (days)	Indicate a reason for each hospital visit
Emergency Department*						
Short-term hospital visit (<24 hours admission)						
Hospitalization in general ward#						
Hospitalization in intensive/critical care						
Mechanical ventilation use						

*Please count Emergency Department visits only if the visit did not result in a hospital admission.

#Please capture type of ward and length of stay in each ward.

4. Institutional care admission(s) other than hospital

Please indicate if there has been any need for admission for care in a long-term facility, since onset of the confirmed COVID-19 episode.

Yes: ____
No: ____

	No	Yes	If yes, specify number of admissions	Specify number of admissions related to COVID-19 or its complications	Specify the length of each stay (days)	Indicate a reason for each institutional care admission
Long-term facilities						
Rehabilitation facility						
Supplemental oxygen						

10.7. Appendix 7: Medically-attended COVID-19 (MA-COV) Form

Section 1: To be completed in all healthcare settings[a] (eg, family doctor, nurse practitioner, outpatient clinic, emergency department visits, and hospitalizations).

Participant ID (to be completed by study staff):
Date of visit:
Name and role of healthcare professional completing form:
Contact details for healthcare professional:

DIAGNOSIS/DIAGNOSES
Please list diagnosis/ diagnoses made during the patient's clinical interactions at this facility.

MEDICATIONS
Please list any new medications prescribed or changes in medication dosing.

CLINICAL NARRATIVE INCLUDING COURSE OF INFECTION

COVID-19 DIAGNOSTIC TEST

Was a COVID-19 diagnostic test performed? ☐ **Yes** ☐ **No**
If 'yes' selected, please fill out remaining questions below

Specify diagnostic method: _____

Specify test name and manufacturer: _____

Date performed: _____

Type of sample taken:_

☐ **Nasal swab sample** ☐ **Saliva sample**

☐ **Sputum sample** ☐ **Other (specify):** _____

Specify results: _____

[a] The MA-COV form should be completed by the medical care provider or study site personnel during medical visits for COVID-19 or COVID-19 complications.

VITAL SIGNS

Has vital sign assessment been performed?

□ **Yes** □ **No**

Temperature (ºC/ºF): _____

Respiratory rate: _____

Pulse: _____

Systolic and Diastolic Blood Pressure: _____

Oxygen saturation: _____

- **Does the subject have a clinically abnormal oxygen saturation?**
  □ **Yes** □ **No**
- **If yes, is the oxygen saturation adjusted for altitude per the investigator judgement:**
  □ **≤93%** □ **>93%**

DIAGNOSTIC TESTING

Was a peak flow measurement made? □ **Yes** □ **No**

If yes, please indicate date performed: _____ _____

Peak flow (L/min): _____

Was a chest X-ray and/or CT performed? □ **Yes** □ **No**

If yes, please indicate date performed: _____

What percentage of the lung was involved? _____

Was an arterial blood gas measured? □ **Yes** □ **No**

If yes, please indicate date performed: _____

Specify results:
pH: _____; pCO_2 (mmHg): _____; pO_2 (mmHg): _____; HCO_3 (mEq/L): _____; O_2 saturation (%):_____

Were additional diagnostic tests performed? □ **Yes** □ **No**

If yes, please specify diagnostic method:

Date performed: _____

Specify results: _____

SIGNS AND SYMPTOMS

In case the severity and/or start and/or end date of any of the experienced signs and symptoms are known, please indicate.

Did the patient experience any of these events, signs or symptoms?

- **Clinical signs at rest indicative of severe systemic illness (respiratory rate ≥30 breaths/minute or heart rate ≥125 beats/minute or SpO2 ≤93% on room air at sea level[a] or PaO2/FiO2 <300 mmHg)**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Respiratory failure requiring high-flow oxygen, non-invasive ventilation, mechanical ventilation, or ECMO**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Respiratory rate ≥20 breaths/minute**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Shortness of breath**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Heart rate ≥90 beats/minute**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Shock (systolic blood pressure <90 mm Hg, or diastolic blood pressure <60 mm Hg or requiring vasopressors)**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Radiologic evidence of DVT**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**
- **Significant acute renal or hepatic dysfunction**
  - **□ Yes □ No**
  - **Severity: □ Mild □ Moderate □ Severe**
  - **□ Start date: _____ □ End date: _____**

[a] SpO_2 criteria will be adjusted according to altitude per investigator judgment.

- **Hyperinflammatory Syndrome**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: □ End date:

- **Symptoms or signs of stroke**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Numbness, tingling, or weakness face or limbs**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Difficulty speaking or forming speech**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Difficulty understanding speech**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Feelings of confusion**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Clinical or radiological evidence of pneumonia**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Fever (≥38.0°C or ≥100.4°F)**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Shaking chills or rigors**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Cough**
  □ Yes □ No
  Severity: □ Mild □ Moderate □ Severe
  □ Start date: _____ □ End date: _____

- **Sore throat**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Malaise**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Headache**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Myalgia**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Gastrointestinal symptoms**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Chilblains/pernio (red or bruised looking feet or toes)**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____
- **Anosmia (olfactory or taste disorders)**
  - □ Yes □ No
  - Severity: □ Mild □ Moderate □ Severe
  - □ Start date: _____ □ End date: _____

MANAGEMENT

ANY TYPE OF MANAGEMENT OTHER THAN MEDICATION? □ Yes □ No

If yes, please specify:

- **Nebulizer treatments**
  - □ Yes □ No
- **IV fluids**
  - □ Yes □ No
- **Intubation**
  - □ Yes □ No

Section 2: COVID-19-related Procedures completed during the event.

SUPPLEMENTAL OXYGEN

Was supplemental oxygen administered? ☐ Yes ☐ No

If 'yes' selected, please fill out remaining questions in this section.

Type of supplemental oxygen administration:

- ☐ Invasive Mechanical Ventilation
- ☐ Non-Invasive Mechanical Ventilation
- ☐ Nasal Cannula
- ☐ Nonrebreathing Face Mask with Reservoir and One-Way Valve
- ☐ Venturi Mask
- ☐ Simple Face Mask
- ☐ Reservoir Cannulas
- ☐ Other: _____

If invasive mechanical ventilation, specify:

- ☐ Through endotracheal tube
- ☐ Through tracheostomy tube

If non-invasive mechanical ventilation, specify:

- ☐ Continuous positive airway pressure
- ☐ Bilevel positive airway pressure

Oxygen concentration and units: _____

Start date and time: _____

End date and time (if applicable): _____

Has supplemental oxygen administration returned to that level provided prior to the current respiratory illness?

☐ Yes ☐ No

DIALYSIS

Was dialysis performed? ☐ Yes ☐ No

If yes, please specify: _____

ANY OTHER PROCEDURES PERFORMED

Were any other procedures for COVID-19 performed? ☐ Yes ☐ No

If yes, please specify:

- **Procedure:**_____
- **Reason performed:** _____

10.8. Appendix 8: Toxicity Grading Scale

Adapted from the FDA Guidance document "Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials" (September 2007) (US DHHS 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; 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[#]	25 – 50 mm	51 – 100 mm	> 100 mm	Hospitalization; Necrosis or exfoliative dermatitis
Swelling[#]	25 – 50 mm	51 – 100 mm	> 100 mm	Hospitalization; Necrosis

[#] Revised by the sponsor.

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 > 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 – 155	> 155	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.
** 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.
[#] Revised by the sponsor.

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; 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 > 800 gms/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
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

[#] Revised by the sponsor.

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	No interference with activity	Some interference with activity not requiring medical intervention	Prevents daily activity and requires medical intervention	Hospitalization[#]

[#] Revised by the sponsor.

B: Tables for Laboratory Abnormalities

Laboratory tests may be performed during routine medical care and assessment of AEs or other medical events based on the investigator's judgment.

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

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)**
Sodium – Hyponatremia mEq/L	132 – 134	130 – 131	125 – 129	< 125
Sodium – Hypernatremia mEq/L	144 – 145	146 – 147	148 – 150	> 150
Potassium – Hyperkalemia mEq/L	5.1 – 5.2	5.3 – 5.4	5.5 – 5.6	> 5.6
Potassium – Hypokalemia mEq/L	3.5 – 3.6	3.3 – 3.4	3.1 – 3.2	< 3.1
Glucose – Hypoglycemia mg/dL	65 – 69	55 – 64	45 – 54	< 45
Glucose – Hyperglycemia Fasting mg/dL Random – mg/dL	 100 – 110 110 – 125	 111 – 125 126 – 200	 >125 >200	 Insulin requirements or hyperosmolar coma
Blood Urea Nitrogen (BUN) mg/dL	23 – 26	27 – 31	> 31	Requires dialysis
Creatinine – mg/dL	1.5 – 1.7	1.8 – 2.0	2.1 – 2.5	> 2.5 or requires dialysis
Calcium – hypocalcemia mg/dL	8.0 – 8.4	7.5 – 7.9	7.0 – 7.4	< 7.0
Calcium – hypercalcemia mg/dL	10.5 – 11.0	11.1 – 11.5	11.6 – 12.0	> 12.0
Magnesium – hypomagnesemia mg/dL	1.3 – 1.5	1.1 – 1.2	0.9 – 1.0	< 0.9
Phosphorous – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	1.6 – 1.9	< 1.6
Creatine phosphokinase (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

Serum *	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**	**Potentially Life Threatening (Grade 4)****
Cholesterol	201 – 210	211 – 225	> 226	---
Pancreatic enzymes – amylase, lipase	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.1 – 5.0 x ULN	> 5.0 x ULN

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

** The clinical signs or symptoms associated with laboratory abnormalities might result in characterization of the laboratory abnormalities as potentially life-threatening (Grade 4). For example. a low sodium value that falls within a Grade 3 parameter (125-129 mEq/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/mm3	10,800 – 15,000	15,001 – 20,000	20,001 – 25, 000	> 25,000
WBC Decrease - cell/mm3	2,500 – 3,500	1,500 – 2,499	1,000 – 1,499	< 1,000
Lymphocytes Decrease - cell/mm3	750 – 1,000	500 – 749	250 – 499	< 250
Neutrophils Decrease - cell/mm3	1,500 – 2,000	1,000 – 1,499	500 – 999	< 500
Eosinophils - cell/mm3	650 – 1500	1501 - 5000	> 5000	Hypereosinophilic
Platelets Decreased - cell/mm3	125,000 – 140,000	100,000 – 124,000	25,000 – 99,000	< 25,000
PT – increase by factor (prothrombin time)	1.0 – 1.10 x ULN**	1.11 – 1.20 x ULN	1.21 – 1.25 x ULN	> 1.25 ULN
PTT – increase by factor (partial thromboplastin time)	1.0 – 1.2 x ULN	1.21 – 1.4 x ULN	1.41 – 1.5 x ULN	> 1.5 x ULN
Fibrinogen increase - mg/dL	400 – 500	501 – 600	> 600	--
Fibrinogen decrease - mg/dL	150 – 200	125 – 149	100 – 124	< 100 or associated with gross bleeding or disseminated intravascular coagulation (DIC)

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

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

Urine *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life-Threatening (Grade 4)
Protein	Trace	1+	2+	Hospitalization or dialysis
Glucose	Trace	1+	2+	Hospitalization for hyperglycemia
Blood (microscopic) – red blood cells per high power field (rbc/hpf)	1 - 10	11 – 50	> 50 and/or gross blood	Hospitalization or packed red blood cells (PRBC) transfusion

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

10.9. Appendix 9: Summary of Guidance from CDC Website[a] on Underlying Medical Conditions That Lead or Might Lead to Increased Risk for Severe Illness From COVID-19

Adults of any age with **certain underlying medical conditions** are at increased risk for severe illness from COVID-19: Severe illness from COVID-19 is defined as hospitalization, admission to the ICU, intubation or mechanical ventilation, or death.

Adults of any age with the following conditions can be more likely to become severely ill from COVID-19[b]:

- Cancer
- Chronic kidney disease
- Chronic lung diseases, including COPD (chronic obstructive pulmonary disease), asthma (moderate-to-severe), interstitial lung disease, cystic fibrosis, and pulmonary hypertension
- Dementia or other neurological conditions
- Diabetes (Type 1 and Type 2)
- Down syndrome
- Heart conditions, such as heart failure, coronary artery disease, cardiomyopathies, or hypertension
- HIV infection
- Immunocompromised state (weakened immune system)
- Liver disease
- Overweight (BMI of >25 kg/m^2 but <30 kg/m^2), obesity (BMI of ≥30 kg/m^2 but <40 kg/m^2), or severe obesity (BMI ≥40 kg/m^2)
- Pregnancy
- Sickle cell disease or thalassemia
- Smoking (current and former)
- Solid organ or blood stem cell transplant
- Stroke or cerebrovascular disease, which affects blood flow to the brain
- Substance use disorder

[a] Source: https://www.cdc.gov/coronavirus/2019-ncov/need-extra-precautions/people-with-medical-conditions.html?CDC AA refVal=https%3A%2F%2Fwww.cdc.gov%2 Fcoronavirus% 2F2019-ncov%2Fneed-extra-precautions%2Fgroups-at-higher-risk.html. Updated 29 March 2021. Accessed: 6 April 2021.

[b] The list of underlying medical conditions is not exhaustive and only includes conditions with sufficient evidence to draw conclusions.

10.10. Appendix 10: Thrombotic Events to be Reported as AESIs

At the time of protocol writing, the list of thrombotic events to be reported to the sponsor as AESIs is provided below. Further guidance may become available on thrombotic events of interest.

- MedDRA PTs for large vessel thrombosis and embolism:
  - Aortic embolus, aortic thrombosis, aseptic cavernous sinus thrombosis, brain stem embolism, brain stem thrombosis, carotid arterial embolus, carotid artery thrombosis, cavernous sinus thrombosis, cerebral artery thrombosis, cerebral venous sinus thrombosis, cerebral venous thrombosis, superior sagittal sinus thrombosis, transverse sinus thrombosis, mesenteric artery embolism, mesenteric artery thrombosis, mesenteric vein thrombosis, splenic artery thrombosis, splenic embolism, splenic thrombosis, thrombosis mesenteric vessel, visceral venous thrombosis, hepatic artery embolism, hepatic artery thrombosis, hepatic vein embolism, hepatic vein thrombosis, portal vein embolism, portal vein thrombosis, portosplenomesenteric venous thrombosis, splenic vein thrombosis, spontaneous heparin-induced thrombocytopenia syndrome, femoral artery embolism, iliac artery embolism, jugular vein embolism, jugular vein thrombosis, subclavian artery embolism, subclavian vein thrombosis, obstetrical pulmonary embolism, pulmonary artery thrombosis, pulmonary thrombosis, pulmonary venous thrombosis, renal artery thrombosis, renal embolism, renal vein embolism, renal vein thrombosis, brachiocephalic vein thrombosis, vena cava embolism, vena cava thrombosis, truncus coeliacus thrombosis

- MedDRA PTs for more common thrombotic events:
  - Axillary vein thrombosis, deep vein thrombosis, pulmonary embolism, MedDRA PTs for acute myocardial infarction*, MedDRA PTs for stroke*

Source: Shimabukuro T. CDC COVID-19 Vaccine Task Force. Thrombosis with thrombocytopenia syndrome (TTS) following Janssen COVID-19 vaccine. Advisory Committee on Immunization Practices (ACIP). April 23, 2021. https://www.cdc.gov/vaccines/acip/meetings/slides-2021-04-23.html.

*Vaccine Adverse Event Reporting System (VAERS) Standard Operating Procedures for COVID-19 (as of 29 January 2021) https://www.cdc.gov/vaccinesafety/pdf/VAERS-v2-SOP.pdf

10.11. Appendix 11: TTS AESI Form

The form below represents the type of information that may be collected in case of a suspected AESI in order to help adjudicate whether the event is a case of TTS. Additional data may be requested by the sponsor for investigation of the event.

Topic of Interest Questionnaire (TOIQ) for Venous Thromboembolism (VTE)

Manufacturer Control Number: Date of Report: [dd-MMM-yyyy]
Product Generic (TRADE) Name:

1. Adverse Event Description

Patient's clinical signs and symptoms

☐ Leg/Calf Oedema	☐ Pain in Leg/Calf	☐ Haemoptysis
☐ Dyspnoea	☐ Chest Pain/Discomfort	☐ Syncope
☐ Tachypnoea	☐ Tachycardia	☐ Cough
☐ Other symptoms:		

Was patient on VTE prophylaxis? ☐ No ☐ Yes, details:

2. Medical History and Concurrent Conditions

Provide details:

Is the patient overweight or obese? If available, please provide height/weight and BMI	☐ No ☐ Yes
Does the patient have a sedentary lifestyle?	☐ No ☐ Yes – details:
Has the subject been in a sitting position for long periods of time prior to the event?	
Is there a history of smoking?	☐ No ☐ Yes – details:
Is there a history of cancer?	☐ No ☐ Yes – details:
Any past medical history of autoimmune disease (i.e., collagen-vascular disease, inflammatory bowel disease) or myeloproliferative disease?	☐ No ☐ Yes – details:
Does the subject have a history of a previous clotting disorder or a diagnosis of a hypercoagulable state?	☐ No ☐ Yes – details:
Is there a prior history of varicose veins, trauma to the involved leg or pelvis, DVT/PE/VTE?	☐ No ☐ Yes – details:
Is there a history of blood transfusion?	☐ No ☐ Yes – details:
Was the patient (female) pregnant at the time of event?	☐ No ☐ Yes – details:
Is there a history of cardiovascular disorder?	☐ No ☐ Yes – details:
Is there a history of organ transplantation?	☐ No ☐ Yes – details:

Genetic risk factors:

☐ Dysfibrinogenemia	☐ Antiphospholipid syndrome	☐ Factor V Leiden mutation
☐ Protein C or S deficiency	☐ Elevated factor VIII levels	☐ Anti-thrombin deficiency
☐ Hyperhomocysteinemia	☐ Prothrombin gene mutation	☐ Blood-clotting disorder
☐ Thrombophilia		

Acquired risk factors:

☐ Reduced mobility (paralysis, paresis, travel etc.)	☐ Recent trauma
☐ Dehydration	☐ Recent discontinuation of warfarin
☐ Recent surgery	☐ Hormone replacement therapy
☐ Concomitant oral contraceptive use	☐ Indwelling central venous catheters
☐ Phlebitis	☐ Lupus

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TOIQ for Venous Thromboembolism (VTE)

MCN:

☐ Inflammatory bowel disease
☐ Diabetes mellitus
☐ Hypertension
☐ Myeloproliferative disorders
☐ Hyperlipidemia
☐ Other significant medical co-morbidities or risk factors for DVT, specify:

If yes to any of the above, provide details:

Provide Well's score, if calculated:

3. **Relevant results of diagnostic tests including laboratory tests, imaging, biopsies, etc.** (Note the levels/conclusion, date performed, normal ranges as well as any other details. **Alternatively, attach full reports of the diagnostic tests.**)

Diagnostic Test	Results at baseline or prior to use of product (Include date and value/details)	Test results after use of product (Include date and value/details)
Clotting Profile (PT, aPTT- prior to anticoagulation treatment)		
Thrombin time (Bovine) Plasma		
Fibrinogen levels		
D-Dimer levels		
Factor V Leiden		
Prothrombin		
Antithrombin activity		
Protein C activity		
Protein S activity		
Homocystein levels		
Dilute Russells Viper Venom Time (DRVVT), Plasma		
Activated Protein C Resistance V (APCRV), Plasma		
Thrombophilia interpretation		
Anticardiolipin antibodies (IgG and IgM) or beta-2 glycoproteins antibodies		
Antiphospholipid antibodies (IgG and IgM)		
Lupus anticoagulant		
Heparin antibodies CBC with smear (microscopic evaluation)		
ESR		
ANA and ANCA		
IL6 levels		

MCN:

C-reactive protein		
ADAMTS13 Activity Assay		
Ceruloplasmin		
Direct Coombs test		
Complement C3, C4		
MethylenetetraHydrofolate reductase gene mutation		
Prothrombin gene mutation (G20210A)		
Occult blood in stool		
COVID-19 test		
Troponins		
Brain Natriuretic Peptide		
Arterial Blood Gas		
Chest X-Ray		
Electrocardiography		
Echocardiography		
Duplex Ultrasonography		
MRI		
CT		
Contrast Venography		
Pulmonary Angiography		
Ventilation-Perfusion Scanning		

Provide details of any additional diagnostic results:

10.12. Appendix 12: Protocol Amendment History

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

Amendment 5 (17 Decemeber 2021)

Overall Rationale for the Amendment: This amendment is written primarily to clarify procedures in the Schedule of Activities. All changes are listed below, including the rationale for each change and a list of all applicable sections.

Section number and Name	Description of Change	Brief Rationale
1.3 Schedule of Activities	Text added to Schedule of Activities to denote 'no subset' for N-serology and humoral immunity samples at study visit 7A.	To clarify that these samples are collected from the 'no subset' participants only.
1.3 Schedule of Activities	Correction of visit timing of Visit 7A.	Visit timing should read 'Vac +119 d'.
1.3 Schedule of Activities	Vital sign measurements added at Visit 10 for 'All' participants.	Vital sign measurements should be scheduled for Visit 10.
1.1 Synopsis 9.5 Planned Analysis	'Study Responsible Physician/Study Responsible Scientist' removed from text on data collection, data management, and safety monitoring.	SRS will no longer be directly involved in data collection, data management and safety monitoring. SRS will be unblinded to allow review of primary analysis.
Throughout the protocol	Minor formatting changes were made.	Minor errors were noted

Amendment 4 (30 November 2021)

Overall Rationale for the Amendment: This amendment is written to add a blood draw for humoral immunogenicity at the 4-month (Day 120) timepoint, to allow assessment of the trend for duration of the immune response (i.e; kinetic curve). This and other changes are listed below, including the rationale for each change and a list of all applicable sections.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 1.3 Schedule of Activities 4.1 Overall Design 8. STUDY ASSESSMENTS AND PROCEDURES 10.2 APPENDIX 2	A Day 120 blood draw has been added (for humoral immunity assessments) for participants not assigned to any subset.	To allow assessment of the durability of the immune response at 4 months post-boost, to in order to evaluate the kinetics of antibody responses overtime.
1.3 Schedule of Activities 8. STUDY ASSESSMENTS AND PROCEDURES	Exploratory blood draws at Day 28 and Day 120 have been added for participants at BIDMC.	To allow assessment of in-depth exploratory immunogenicity endpoints at additional timepoints.
6.7 Concomitant Medications	Added requirement for Cohort 2 participants to provide dates of primary vaccination with BNT162b2.	To clarify that collection of primary BNT162b2 vaccination dates is necessary for the proper conduct of the analysis.
7.2 Participant Discontinuation/Withdrawal	Added text to state that participants receiving a Covid-19 booster vaccine off study will be excluded from all formal hypothesis testing (primary objectives).	Off study booster vaccinations will make participants unevaluable in the protocol defined primary endpoints.
Title Page	Study name ‘Amplify’ added to Title page	Study name should have been included on title page.
Throughout the protocol	Minor grammatical, formatting changes and clarifications were made.	Minor errors were noted

Amendment 3 (08 October 2021)

Overall Rationale for the Amendment: This amendment is written to make the planned interim analyses for Cohort 1 optional as study enrollment progress indicates that the timing of interim analysis and primary analysis will be too close in time to make the interim analysis of significant added value to the study and to add a Day 29 phone call for all participants to ensure robust reporting of the unsolicited AEs.

These and other changes made to the clinical protocol of study VAC31518COV2008 are listed below, including the rationale for each change and a list of all applicable sections.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 4.1 Overall Design 9.1 Statistical Hypotheses 9.2 Sample Size Determination 9.4 Statistical Analyses 9.4.2 Primary Endpoints 9.5 Planned Analysis	Text was updated to make the planned interim analysis for Cohort 1 optional.	See overall rationale above
1.3.1 All Participants	Added a Day 29 phone call for all participants.	To ensure robust reporting of the unsolicited AEs in the study.
1.1 Synopsis 9.5 Planned Analysis	For the primary analysis, text for the immunogenicity data was updated to include "S-ELISA and/or MSD, if available at the time of analysis".	To align with the secondary endpoints.
1.1 Synopsis 9.5 Planned Analysis	Text was added to clarify the unblinding of data at the time of primary analysis with respect to each cohort.	Clarification
1.1 Synopsis 1.3.1 All Participants 1.3.3 Participants With COVID-19-like Signs and Symptoms 4.1 Overall Design 8.2.2 Vital Signs	Text was updated to clarify that the preferred position for collection of blood pressure measurements is supine.	Clarification
Throughout the protocol	Minor grammatical, or formatting changes were made and minor clarifications, including alignment across sections, were added.	Minor errors were noted

Amendment 2 (22 September 2021)

Overall Rationale for the Amendment: This amendment is written to address Health Authority (HA) feedback, including, the addition of formal non-inferiority (NI) hypothesis testing for the leading severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) variant of high consequence or concern, as defined by the Centers for Disease Control and Prevention (CDC Aug 2021c), the adjustment of the primary objectives for Cohort 2, which will now compare post-boost neutralizing antibodies with neutralizing antibodies post Pfizer primary vaccine series, the addition of another success criterion for the NI assessments, requiring a geometric mean ratio (GMR; geometric mean titer [GMT] Day 15 post booster/GMT post primary regimen) >0.8 in order to conclude NI, and the extension of the follow-up phase from 6 months to 1 year. In addition, an interim analysis is implemented in this study to formally test Primary Objectives 1a and 1b and the time windows for Visits 5 and 6 have been adjusted.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 4.1 Overall Design 9.1 Statistical Hypotheses 9.2 Sample Size Determination 9.4 Statistical Analyses 9.4.2 Primary Endpoints 9.5 Planned Analysis	When at least 330 participants from Groups 1 to 3 in Cohort 1 have been enrolled, have completed the Day 15 visit and it is estimated that immunogenicity data can be obtained from 110 or more participants in Group 1, an interim analysis will be conducted whereby formal non-inferiority testing of Cohort 1 – Group 1 (Primary Objectives 1a and 1b) will be performed on the available data from the Cohort 1 – Group 1 participants.	To demonstrate NI of the immunogenicity post booster vaccination versus post Ad26.COV2.S primary regimen.
1.1 Synopsis 2 INTRODUCTION 3 OBJECTIVES AND ENDPOINTS 8.1.5 Immunogenicity Assessments 9.1 Statistical Hypotheses 9.4.2 Primary Endpoints 11 REFERENCES	The study endpoints have been revised to include formal hypothesis testing (primary objective/endpoint) for the effectiveness of the Ad26.COV2.S booster vaccination at $5x10^{10}$ vp following a primary regimen of a single dose of Ad26.COV2.S at $5x10^{10}$ vp or a 2-dose Pfizer BNT162b2 regimen against leading SARS-CoV-2 variant of high consequence or concern, as defined by the CDC (CDC, Aug2021c) in the US at the time of the analysis. Additional relevant variants may be analyzed as secondary objective.	HA request to include study endpoints, with formal hypothesis testing, evaluating effectiveness against circulating variants that are relevant based on US epidemiology data.
1.1 Synopsis 1.2 Schema 2 INTRODUCTION 3 OBJECTIVES AND ENDPOINTS 8.1.5 Immunogenicity Assessments 9.1 Statistical Hypotheses 9.3 Analysis Sets 9.4.2 Primary Endpoints	Primary objectives for Cohort 2 have been revised. For the NI assessments neutralizing antibodies post booster will now be compared to neutralizing antibody response post the Pfizer primary vaccination instead of the neutralizing antibody response post a single Ad26.COV2.S ($5x10^{10}$ vp) dose. Serum samples from approximately 300 individuals who received the Pfizer primary vaccine series will be obtained from Biobank or other external studies to perform this analysis.	HA request to include assessment of immune responses after a booster dose of Ad26.COV2.S in participants who received the primary Pfizer vaccine series compared to the responses after the primary Pfizer vaccine series to ensure that post booster responses are at least the response observed post primary vaccination with the Pfizer vaccine.
1.1 Synopsis 3 OBJECTIVES AND ENDPOINTS 9.1 Statistical Hypotheses 9.4.2 Primary Endpoints	An additional pre-specified success criterion for NI hypothesis testing was added. In addition to statistical NI of responder rate and statistical NI of GMTs, an estimated GMR (GMT Day 15 post booster/GMT post primary regimen) of >0.8 is required to conclude NI.	HA recommendation
1.3 Schedule of Activities	The visit window of Visits 5 (Day 15) and 6 (Day 29) were adjusted ±3 days to -3/+7 days for Visit 5 and -3/+10 days for Visit 6.	Learnings from the ongoing COV2008 study have indicated that it is challenging for participants to have the Visits 5 and 6 within the specified time window. Therefore, the windows for these visits were extended. In view of the immunogenicity results observed in Ad26.COV2.S studies, including VAC31518COV1001 and VAC31518COV2001 studies, a broader visit window for Visits 5 and 6 should not impact the immunogenicity assessment.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 1.3.1 All Participants 3 OBJECTIVES AND ENDPOINTS 4.1 Overall Design 4.4 End of Study Definition 8 STUDY ASSESSMENTS AND PROCEDURES	The follow-up in the study for safety and immunogenicity has been extended to 12 months post booster.	To collect extended safety data and to collect data on durability of immune response
5.1 Inclusion Criteria	A time window of maximum -20 days was added to the allowed time interval of ≥ 6 months between the primary vaccine regimen and Ad26.COV2.S booster.	To facilitate recruitment
1.3.2 Participants with a Suspected AESI	It is clarified that if an AESI is reported to the investigator more than 28 days after the onset of the event, the Day AESI 29 visit (28 days post the onset of the event) becomes obsolete and does not need to be performed.	Clarification
1.1 Synopsis 9.4.2 Primary Endpoints 11 REFERENCES	It is clarified that for the NI analysis on responder rate the Agresti-Min method is chosen to estimate the difference in proportions and its CI because of its well-behaved coverage probability (CP) properties compared to other methods for the analysis of matched pairs data (Reed 2009). Coverage probability is generally used to evaluate (1 – α) confidence intervals where α is the Type I error rate.	Clarification
10.2 Appendix 2: Clinical Laboratory Tests	It has been added that, as part of investigation of any AESI, samples from appropriate controls (from vaccinated participants who did not experience an AESI) within the study may be used for coagulation-related assays.	Clarification
2.3.1 Risks Related to Study Participation	Text has been added regarding the increased risk of Guillain-Barre Syndrome (GBS) following use of the Ad26.COV2.S vaccine.	Based on the emerging data following use of the Ad26.COV2.S vaccine, GBS has been identified as an adverse drug reaction for the use of Ad26.COV2.S vaccine.
1.1 Synopsis 11 REFERENCES	The reference to the Brighton Collaboration case definition of thrombotic events and thrombocytopenia was updated.	Update
8 STUDY ASSESSMENTS AND PROCEDURES	Blood volumes have been corrected and it is clarified that for participants at the BIDMC site, up to an additional 50 mL of blood is taken per blood draw (and not in total) for experimental research amounting to an additional total approximate blood volume of 200 mL.	Correction and clarification
1.3.1 All Participants	It is clarified that the e-diary review at Day 8 is only applicable for participants in blood collection Subset 1. The e-diary will be reviewed at the Day 15 visit for all	Clarification

Section number and Name	Description of Change	Brief Rationale
	participants, including repeated review for the participants in Subset 1.	
1.3.2 Participants with a Suspected AESI	It is clarified that also in the event of thrombocytopenia, laboratory assessments (to be performed locally) are required to facilitate diagnosis and determine treatment options, including but not limited to platelet count and anti-PF4 tests.	Clarification
Throughout the protocol	Minor grammatical, formatting, or spelling changes were made and minor clarifications, including alignment across sections, were added.	Minor errors were noted

Amendment 1 (16 August 2021)

Overall Rationale for the Amendment: This amendment is written to address HA feedback, including the increase of the sample size of study groups of interest and adjustment of the assumption in the sample size calculation for the non-inferiority comparisons of a seroresponse rate of 95% post booster versus 90% post primary vaccination and a geometric mean titer (GMT) ratio of 1.5 for the booster to using the conventional assumption of no difference between the virus neutralization assay (VNA) data post booster and post primary vaccination.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 1.2 Schema 4.1 Overall Design 8 STUDY ASSESSMENTS AND PROCEDURES 9.2 Sample Size Determination 9.4.3 Secondary Endpoint(s)	The sample size of study groups 1, 2, 4 and 5, including a booster dose at the $5x10^{10}$ vp or $2.5x10^{10}$ vp dose level was increased to ~330 participants per study group. In each of the cohorts, participants will initially be randomized in a 1:1:1 ratio into 3 groups to receive a 1-dose booster vaccination regimen with Ad26.COV2.S until group 3 (Cohort 1) or group 6 (Cohort 2) is fully enrolled. Thereafter, randomization will continue in a 1:1 ratio in groups 1 and 2 (Cohort 1) or groups 4 and 5 (Cohort 2). The size of the subsets (subsets 1 to 4) for humoral immunogenicity at intermediate timepoints and cellular immunogenicity remains unchanged. The first ~330 randomized participants per cohort will be assigned to 1 of the 4 blood collection subsets. Once the subsets are enrolled, the subsequent participants will not be in a subset. They will only have the immunogenicity samples taken at Day 1, Day 15 and Day 181.	To increase the safety database of the final chosen booster dose level for each primary series cohort (either 1 dose of Ad26.COV2.S or 2 doses of BNT162b2) to allow for potential assessment of safety events that may occur at a rate of 1% and to accommodate for the revised assumption of no difference between the VNA data post booster and post primary vaccination in the sample size calculation for the non-inferiority comparisons. (see description of change in next row)
1.1 Synopsis 9.2 Sample Size Determination	The assumptions for the non-inferiority comparison of a seroresponse rate of 95% post booster versus 90% post primary vaccination and a GMT ratio (Day 15 post-booster/Day 29 post primary regimen) of 1.5 have been revised to assume no difference between VNA data post booster and post primary vaccination, ie, 90% seroresponse rate post booster and post primary vaccination and a GMT ratio of 1.0.	Upon recommendation of the Health Authority to use the conventional assumption of no difference between the VNA data post booster and post primary vaccination in the sample size calculation for the non-inferiority comparisons.

Section number and Name	Description of Change	Brief Rationale
1.1 Synopsis 9.4.2 Primary Endpoints	It is clarified that the non-inferiority analysis on responder rate will be based on Agresti-Min (Agresti 2005) method to estimate the difference in proportions and its confidence interval.	Clarification
1.1 Synopsis 1.3 Schedule of Activities 3 OBJECTIVES AND ENDPOINTS 8 STUDY ASSESSMENTS AND PROCEDURES	It is clarified that in-depth humoral and cellular immunogenicity assessments may be performed on participants enrolled at the BIDMC site **and in a subset of participants enrolled at other sites**. It is also clarified that for participants enrolled at BIDMC, additional blood draws are required for this exploratory analysis at Day 1, Day 15 and Day 181.	Clarification
Throughout the protocol	Minor grammatical, formatting, or spelling 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 vaccine, 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 Vaccines & Prevention B.V.

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 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	21-Apr-2022 03:56:08 (GMT)	Document Approval