DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

STATISTICAL ANALYSIS PLAN	
Study Code	D7220C00001
Edition Number	5.0
Date	23-Sep-2022

A Phase II/III Partially Double-Blinded, Randomised, Multinational, Active-Controlled Study in Both Previously Vaccinated and Unvaccinated Adults to Determine the Safety and Immunogenicity of AZD2816, a Vaccine for the Prevention of COVID-19 Caused by Variant Strains of SARS-CoV-2 (Master SAP)

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

TABLE OF CONTENTS

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

LIST OF TABLES

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

LIST OF FIGURES

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

LIST OF ABBREVIATIONS

Abbreviation or Specialized Term	Definition
AE	Adverse event
AESI	Adverse event of special interest
BMI	Body mass index
CI	Confidence Interval
COVID-19	Coronavirus 2019
CRF	Case report form
CSP	Clinical study protocol
FAS	Full analysis set
GMT	Geometric mean titre
IMP	Investigational medicinal product
IPD	Important Protocol Deviations
MedDRA	Medical Dictionary for Regulatory Activities
MAAE	Medically attended adverse event
PD	Protocol deviation
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus 2
SD	Standard deviation
SI	Standard International
ULN	Upper limit of normal

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

AMENDMENT HISTORY

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CATEGORY* Change refers to:	**Date**	**Description of change**	**In line with CSP?**	**Rationale**
N/A		Initial approved SAP	N/A	N/A
Primary or secondary endpoints	16 August, 2021	Added non-inferiority comparisons	Yes	Updated for CSP v3.0
Multiple Testing Procedure	16 August, 2021	Added hierarchical approach to control for multiplicity	Yes	Updated for CSP v3.0
Primary or secondary endpoints	20 October, 2021	Remove age cap and revise the primary and key secondary analyses to include historical controls	Yes	Updated for CSP v3.0
Primary or secondary endpoints	17 November, 2021	Added propensity score matching for historical controls; added text to allow for use of an international standard for immunogenicity assays if one becomes available; updated covariate adjustment of time since previous vaccination for model-adjusted immunogenicity values.	Yes	Updated for CSP v4.0 (amendment 3)

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Primary or secondary endpoints	01 February, 2022	Structure current document as master-SAP and add references for sections covered in specific sub-SAPs for participants previously vaccinated with 2 doses of AZD1222, participants previously vaccinated with 2 doses of a mRNA vaccine, and participants previously unvaccinated. Updated hierarchy and definitions of planned primary and key secondary immunogenicity objectives/endpoints in each sub-SAP in accordance with CHMP and MHRA feedback. Include separate hierarchy for participants previously vaccinated with 2 doses of a mRNA vaccine. Add primary safety objective/endpoint for participants previously vaccinated with 2 doses of a mRNA vaccine. Removal of interim analysis 3 Add relevant sensitivity analyses to sub-SAPs to address CHMP and MRHA comments	No	Addressing feedback from CHMP and MHRA. See Section 2 for deviations of planned analyses from CSP.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Statistical analysis method for the primary or secondary endpoints	23 September, 2022	Changed non-inferiority margin in power calculations for seroresponse rate comparisons to -10%, as per CHMP and MHRA comments (analyses using -10% margin were updated previously on 01 February 2022) Removal of primary and secondary analyses (to be completed all with final analysis) Clarified exposure time points for AE analyses Added references to analysis methods Changed CTCAE grading to FDA grading of safety labs	No	Further addressing feedback from CHMP, MHRA, and the DSMB. Further clarifying analysis methods.
Data presentations	23 September, 2022	Subgroup analyses to only be performed where appropriate Removed scatterplots related to safety labs	No	Eliminating displays of little interest

*Pre-specified categories are:

Primary or secondary endpoints; Statistical analysis method for the primary or secondary endpoints; Derivation of primary or secondary endpoints; Multiple Testing Procedure; Data presentations; Other

N/A = Not applicable

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

1 INTRODUCTION

Following feedback from the Committee for Medicinal Products for Human Use (CHMP) it was recommended to have three separate sub Statistical Analysis Plans (SAPs) for those previously vaccinated with two doses of AZD1222, previously vaccinated with two doses of a previous mRNA vaccine, and those previously unvaccinated. These shall henceforth be referred to as the **V1222-SAP**, **VmRNA-SAP**, and **Naïve-SAP** respectively. The current document is the master SAP (master-SAP) which details sections common to all 3 analysis populations.

The purpose of this document is to give details for the statistical analysis of study D7220C00001 supporting the clinical study report. The reader is referred to the Clinical Study Protocol (CSP), amendment 3 dated 11 October 2021 as well as the most recent versions of the Case Report Form (CRF) for details of study conduct and data collection, and the V1222-SAP, the VmRNA-SAP and the Naïve-SAP for details of the analysis principles specific to each sub Statistical Analysis Plan (SAP).

The term IMP (investigational medicinal product) is used throughout this SAP to include both treatment groups (AZD2816 and AZD1222). AZD2816 is specified when referring to participants who received IMP intervention.

1.1 Objectives and Endpoints

1.1.1 Previously unvaccinated cohort receiving 2-dose primary vaccination

Please refer to the Naïve-SAP.

1.1.2 Previously vaccinated cohort receiving 1-dose booster vaccination following two doses of AZD1222.

Please refer to the V1222-SAP.

1.1.3 Previously vaccinated cohort receiving 1-dose booster vaccination following two doses of a mRNA vaccine.

Please refer to the VmRNA-SAP.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

1.2 Study Design

This is a multi-country Phase II/III study to evaluate the safety and immunogenicity of AZD2816 as single-dose vaccination in previously vaccinated adult participants and as a 2-dose primary vaccination in previously unvaccinated adult participants.

A total of approximately 2590 SARS-CoV-2 nucleocapsid seronegative participants that have been screened and judged to be eligible for the study will be enrolled across these 2 populations with the goal of 1300 previously vaccinated participants receiving single-dose vaccination and 1290 unvaccinated participants receiving 2-dose primary vaccination. In addition, seropositive participants will be enrolled (with a cap of 10% of the seronegative population or 259 participants) to support exploratory analysis is these participants. Participants are classified at baseline as either seronegative or seropositive based on result of a lateral flow test.

The enrollment and randomisation strategy are intended to minimize group differences in terms of age, gender and the presence of comorbidities.

In both the single-dose booster treatment regimen and the 2-dose primary vaccination treatment regimen, participants will receive study intervention consisting of intramuscular administration of either AZD1222 (5×10^{10} viral particles) or AZD2816 (5×10^{10} viral particles).

Approximately 700 seronegative participants previously vaccinated with AZD1222 will be randomised 1:1 to receive a single intramuscular dose of either AZD1222 or AZD2816 in a double-blinded fashion.

Approximately 600 seronegative participants previously vaccinated with an approved mRNA based vaccination against the original Wuhan-Hu-1 strain will be randomised 1:1 to receive a single intramuscular dose of AZD2816 or AZD1222 in a double-blinded fashion.

Approximately 1290 seronegative, previously unvaccinated participants will be randomised 5:5:5:2 to receive a 2-dose primary vaccination of the following:

- 2 doses of AZD2816 with a 4-week dosing interval;
- 2 doses of AZD1222 with a 4-week dosing interval;
- 1 dose of AZD1222 followed by 1 dose of AZD2816 with a 4-week dosing interval;
- 2 doses of AZD2816 with a 12-week dosing interval.

The 3 treatments with a 4-week dosing interval will be double-blinded while the treatment with the 12-week interval will be open-label due to the difference in dosing interval.

In addition, a smaller population of seropositive participants (with a cap of 10% of the seronegative population), will be randomised to treatment in a similar manner as above.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Owing to national vaccine rollout in the recruitment countries, including the prioritization of the elderly, it is anticipated that there will be critical differences between the previously vaccinated and previously unvaccinated cohorts that may confound the interpretation of the results. Consequently, the primary and key secondary non-inferiority analyses across these two cohorts will compare the previously vaccinated participants that received a booster dose in this study with a subset of matched participants from the previously unvaccinated participants that received the 2-dose AZD1222 primary vaccine series in the AZD1222 Phase 3 trial, Study D8110C00001.

Immunogenicity (ie, anti-Wuhan-Hu-1 and anti-B.1.351 immune responses including S-binding antibody titres and neutralizing antibody levels [pseudo-neutralization]) will be assessed in serum samples collected pre-dose on the day of each vaccination (baseline levels before vaccination), 14 and 28 days after each vaccination, and 180 days after the last vaccination.

All participants will be given a thermometer, tape measure or ruler, and a proprietary e-diary application designed for use with a smart device with instructions for use. All participants will be asked to report on solicited signs and symptoms for 7 days following vaccination (Days 1-8 for all participants and Days 29-36 for the 4-week dosing interval and Days 85-92 for the 12-week dosing interval). An e-diary will be used to collect information on the timing and severity of the solicited signs and symptoms.

Follow-up visits will take place as per the schedule of assessment within respective windows. All participants will be assessed for local and systemic AE, physical examination, review of e-diaries at these time points as detailed in the schedule of assessment. Blood will also be taken for safety assessments and immunology purposes.

All study participants will be followed for safety for 180 days after administration of their last vaccination dose. In every participant, solicited local and systemic events will be reported for up to 7 days after each dose, all unsolicited AEs will be reported for up to 28 days after each dose, and SAEs and AEs of special interest will be evaluated through study completion (up to 180 days after the last study vaccination).

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Figure 1 Study Design for Unvaccinated Seronegative/Seropositive Participants Receiving a 2-Dose Primary Vaccination

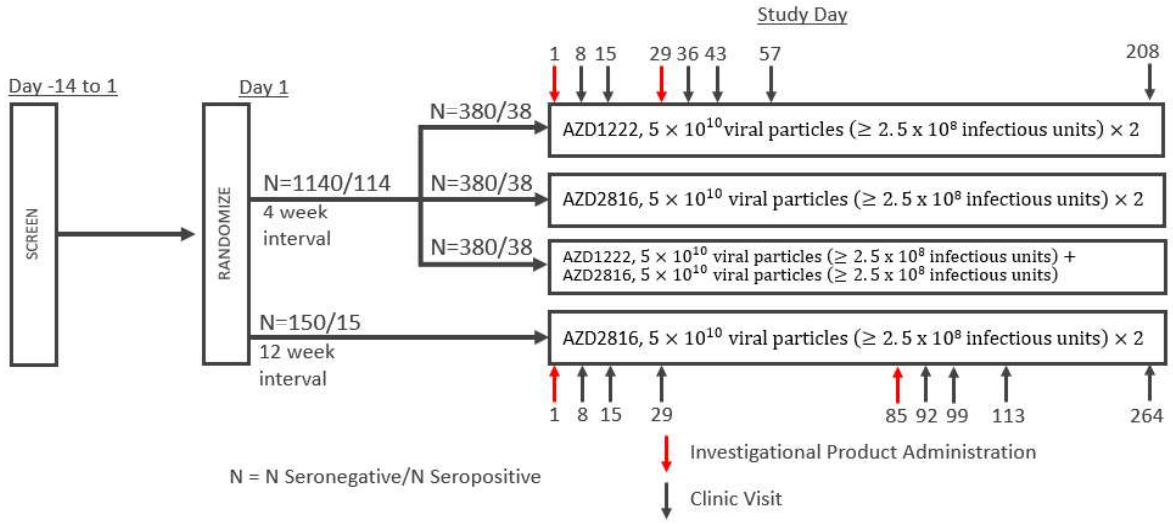
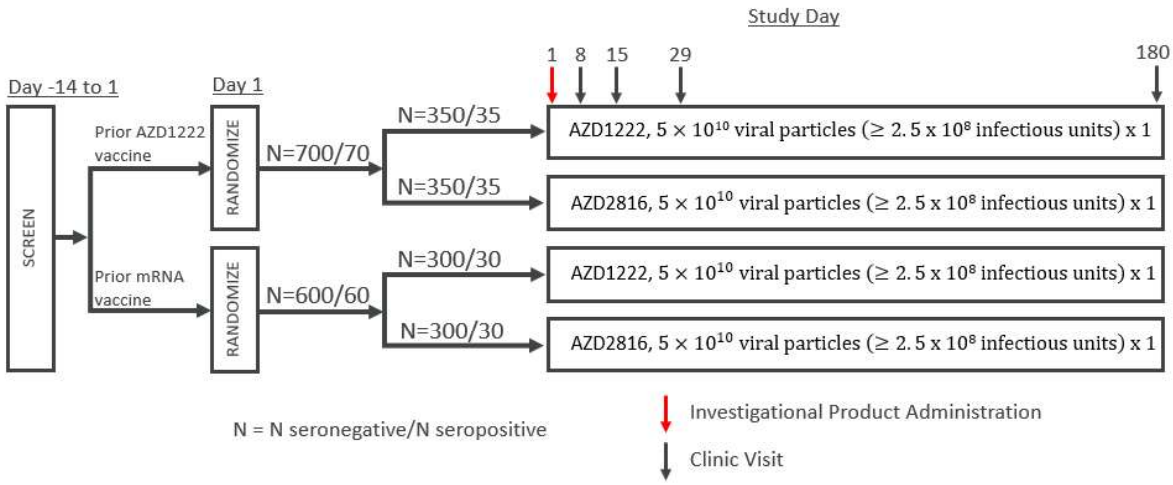


Figure 2 Study Design for Previously Vaccinated Seronegative/Seropositive Participants Receiving a 1-Dose Booster

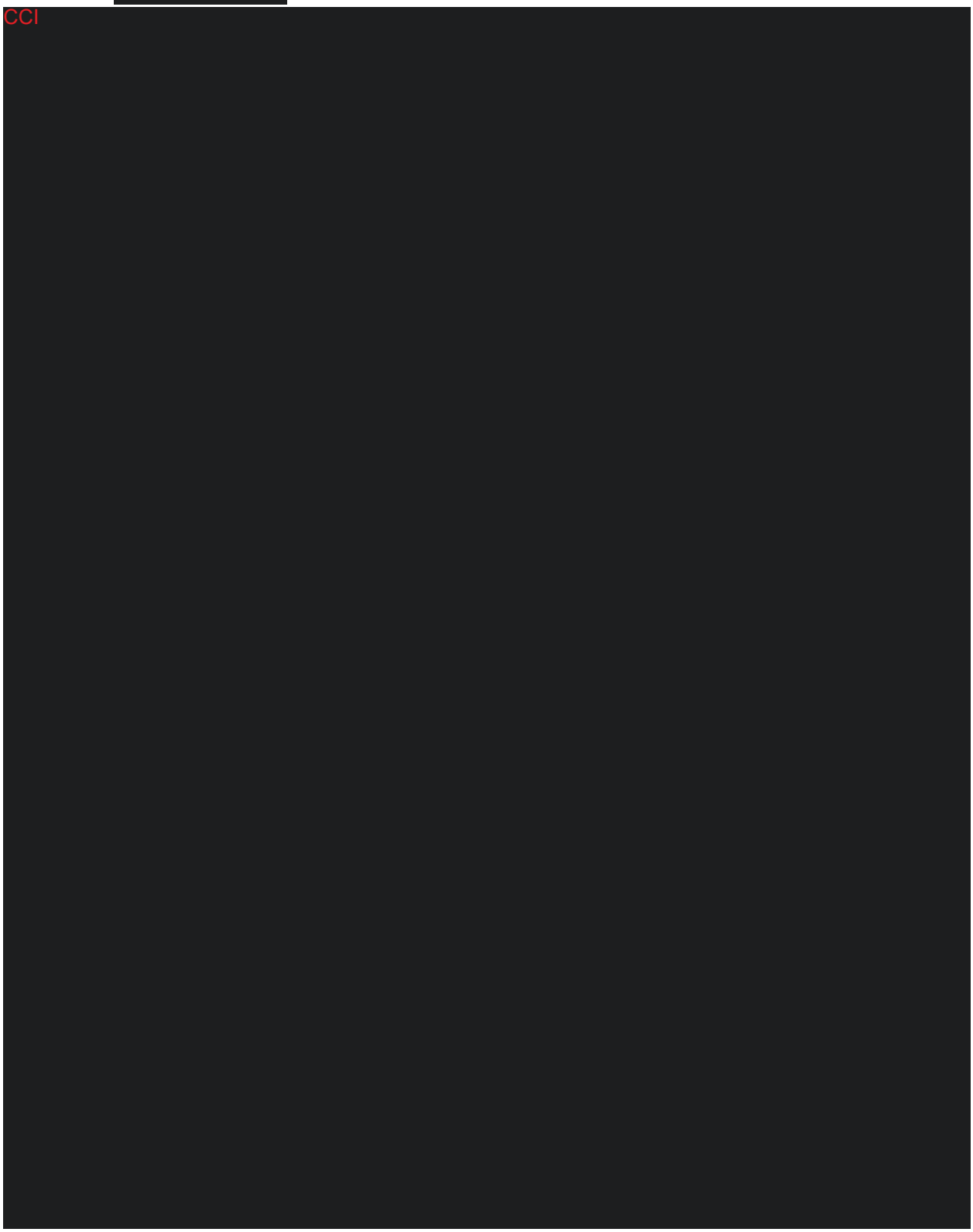


Note: In addition to the approximately 2590 seronegative participants enrolled to support the primary/secondary objectives, seropositive participants will also be enrolled in the study to support exploratory objectives in this population, with a cap of 10% of the planned seronegative participants (ie, a maximum of 259 seropositive participants, bringing total enrollment to 2849).

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

1.3 CCI

CCI

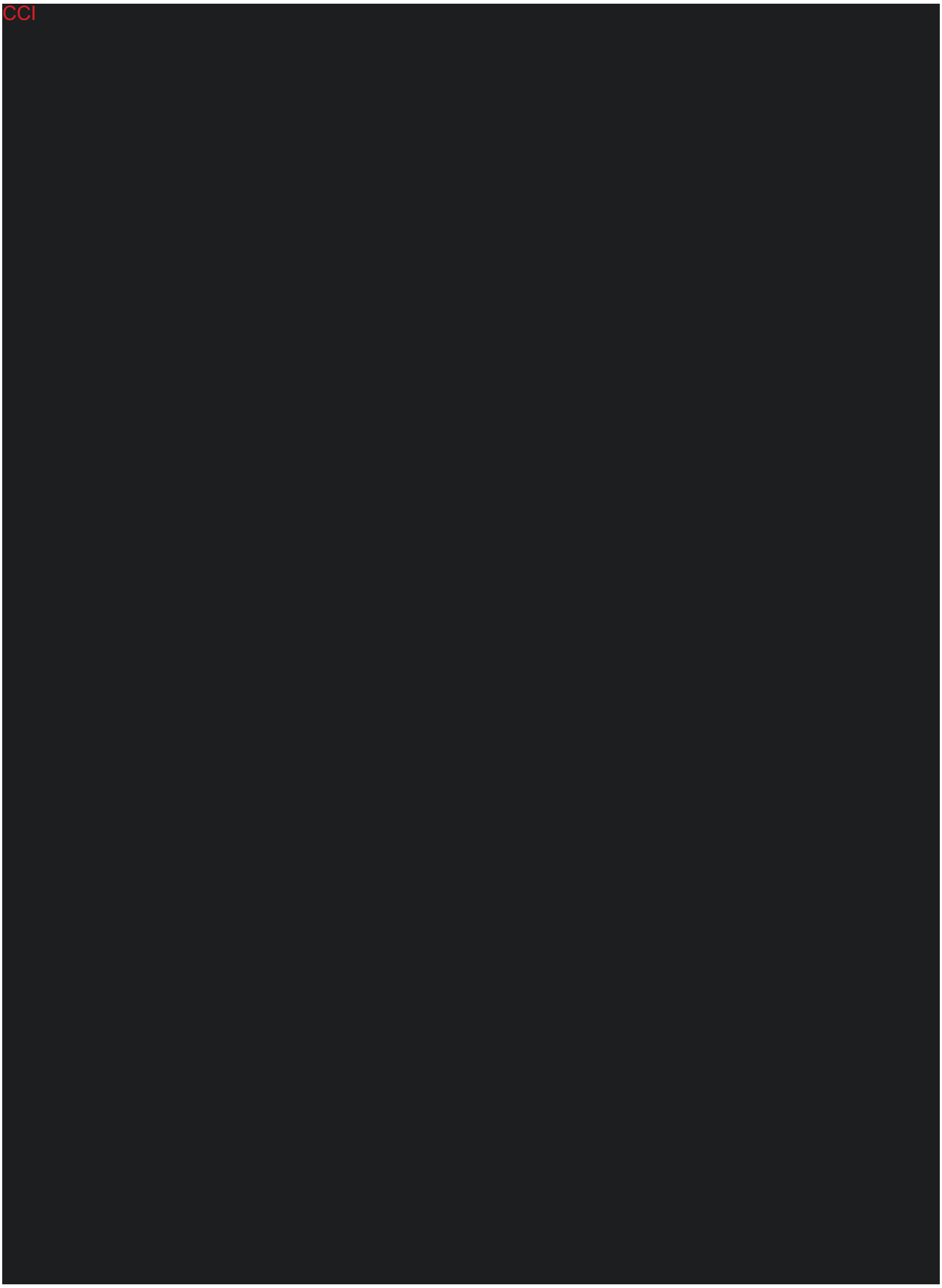


DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI



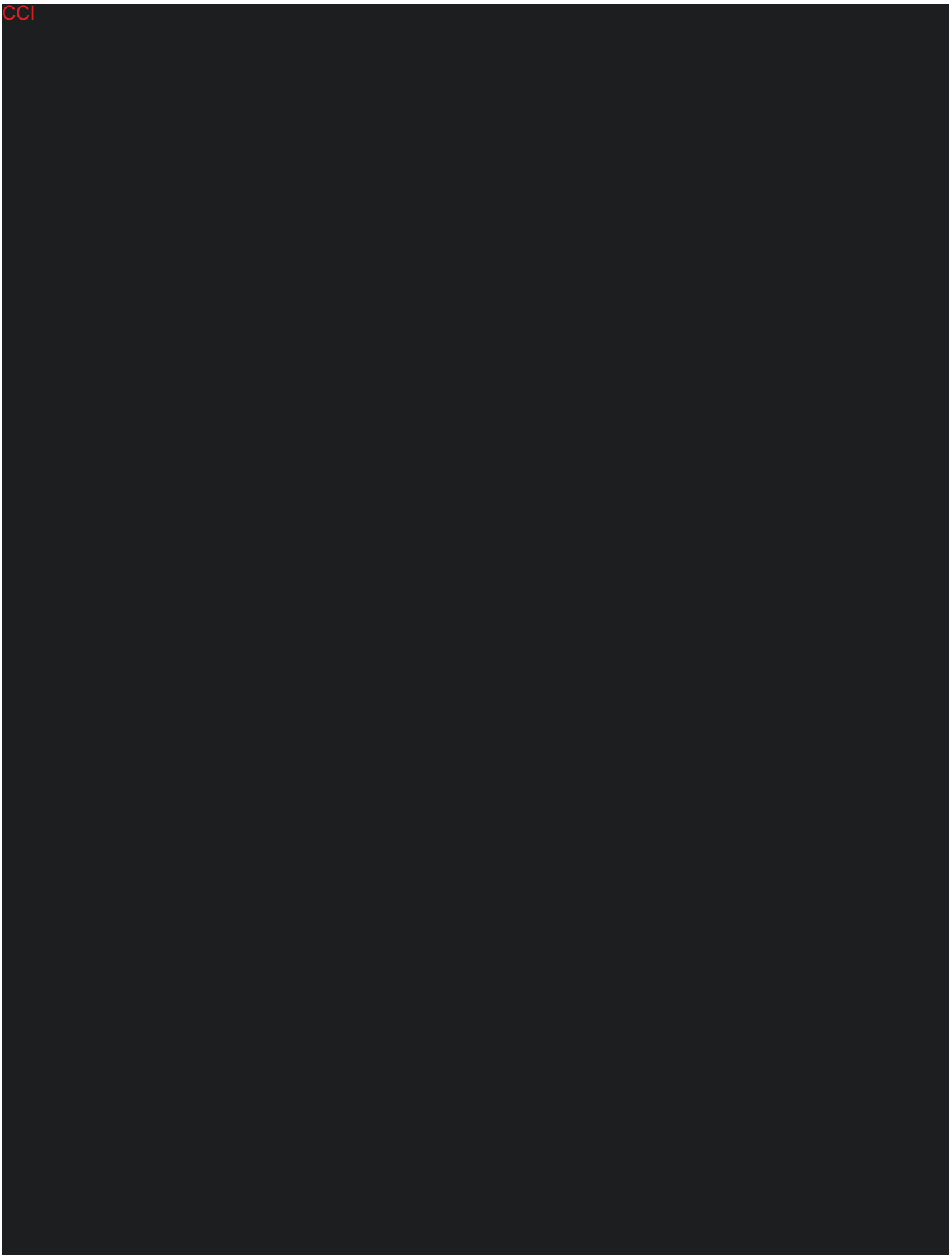
DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI



DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI

[redacted]

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

CCI [redacted]

2 CHANGES TO PROTOCOL PLANNED ANALYSES

Following from feedback received by the CHMP and MHRA the following changes have been made from the analysis originally described in the CSP.

By request of the CHMP, separate SAPs have been created to analyse participants previously vaccinated with AZD1222, participants previously vaccinated with an mRNA vaccination, and previously unvaccinated participants. The current document is the master-SAP. The three sub-SAPs V1222-SAP, VmRNA-SAP and Naïve-SAP have been created to detail the analyses planned for these separate cohorts.

Originally, objectives and endpoints for previously vaccinated participants were pooled for participants previously vaccinated with AZD1222 or a mRNA vaccine, with the primary safety objective, and the hierarchy of primary and key secondary immunogenicity endpoints focused on those previously vaccinated with AZD1222. However, following the creation of three separate sub SAPs requested by the CHMP objectives and endpoints will be constructed separately for participants previously vaccinated with AZD1222, participants previously vaccinated with an mRNA vaccination, and previously unvaccinated participants. Consequently, separate primary safety objectives and hierarchies of primary and key secondary analyses are to be carried out for each of these cohorts.

CCI [redacted]

CCI [redacted] The previous key secondary endpoint 2.4 comparing the humoral immune response elicited by the original Wuhan-Hu-1 strain in participants previously vaccinated with two doses of AZD1222 and a booster dose of AZD1222 against the response elicited by a two dose AZD1222 vaccination administered to unvaccinated participants has been promoted to the current primary endpoint as the CHMP considered this to be the most relevant objective to support the use of AZD1222 as a booster.

In amendment 3 of the CSP it was specified that a list of historical controls in study D8110C00001 was to be selected for immunogenicity comparisons against those who were previously unvaccinated receiving two doses of AZD1222. As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

affected comparative analyses. This affects the previous key secondary endpoint 2.1 for the previously vaccinated cohort.

Owing to national vaccine rollout in the recruitment countries, including prioritization of elderly populations, it is anticipated that there will be critical differences between the previously vaccinated and previously unvaccinated cohorts that may confound the interpretation of results. Therefore, the previous key secondary endpoint 2.1 has been de-prioritised to key secondary endpoint 2.5. Along with the prioritization of previous key secondary endpoint 2.4 to primary, the changes in the hierarchy of endpoints for previously vaccinated cohorts for the new previously vaccinated with AZD1222 cohort is presented in **Table 7**. A similar table format will be considered for the hierarchy of immunogenicity endpoints in participants previously vaccinated with an mRNA vaccine.

Table 7 Changes in Hierarchy of Immunogenicity Endpoints for Previously Vaccinated Cohort

	Previous hierarchy of endpoints for previously vaccinated cohort	**Current hierarchy of endpoints for previously vaccinated participants receiving two doses of AZD1222**
Primary	The neutralizing antibody GMT response against the B.1.351 variant elicited by a AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination administered to previously unvaccinated participants.	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.
Key Secondary 2.1 [a]	The neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants	The neutralizing antibody GMT response against the B.1.351 variant elicited by a AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination administered to previously unvaccinated participants.
Key Secondary 2.2	The neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222	The neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

	booster dose in participants previously vaccinated with AZD1222.	booster dose in participants previously vaccinated with AZD1222.
Key Secondary 2.3	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.
Key Secondary 2.4[b]	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.
Key Secondary 2.5	The neutralizing antibody GMT response against the Wuhan-Hu-1 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.	The neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants

a De-prioritized to key secondary 2.5 in current SAP.
b Prioritized to primary in current SAP

A third interim analysis was planned to be conducted for a blinded sample size re-estimation of naïve cohorts. However, given the challenges to enrolment of naïve participants CCI [redacted], this interim analysis has been removed.

Separate analyses were planned when all data were available for the primary endpoint of the previously unvaccinated 4-week dosing arm cohort, and for the secondary endpoint of the previously unvaccinated 12-week dosing arm cohort. All these analyses are now combined in one after final Database Lock.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

3 DATA ANALYSIS CONSIDERATIONS

3.1 Timing of Analyses

An initial interim analysis will be performed on a subset of previously AZD1222 vaccinated participants that have received a booster dose to consider unblinded sample size adjustment. Full details of this analysis are provided in the Interim Analysis Charter to be finalized prior to any interim analysis.

A second interim analysis will be performed when all previously AZD1222 vaccinated participants have completed their Day 29 visit to support registration of a booster dose. Full details of this analysis are provided in the Interim Analysis Statistical Analysis Plan to be finalized prior to the second interim analysis.

The final analysis will occur when data from all vaccinated participants is available through completion of the last study visit (180 days after the single dose for previously vaccinated participants / 180 days after the second dose for unvaccinated participants).

3.2 Analysis Populations

The following populations are defined:

Table 8 Populations for Analysis

Population/Analysis Set	Description
All participants analysis set	All participants screened for the study, to be used for reporting disposition and screening failures.
Full analysis set	All randomised participants who received study treatment, irrespective of their protocol adherence and continued participation in the study. Participants will be analysed according to their randomised treatment, irrespective of whether or not they have prematurely discontinued, according to the intent-to-treat principle. Participants who withdraw consent or assent to participate in the study will be included up to the date of their study termination.
Safety analysis set	The safety analysis set consists of all participants who have received study treatment. Erroneously-treated participants (eg, those randomised to AZD2816, but were actually given treatment AZD1222) are accounted for in this analysis set by assigning them to the treatment they actually received.
Immunogenicity analysis set	The vaccine immunogenicity analysis set will include all randomised participants, received at least 1 dose of planned study treatment (ie, 1 dose of either AZD2816 or 1 dose of AZD1222), had baseline and post-dose antibody measurements, have at least 1 post-dose quantifiable serum titre, and had no protocol deviations judged to have the potential to interfere with the generation or interpretation of an antibody response. The analyses conducted using this analysis set will be based on the actual treatment received.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Seronegative immunogenicity analysis set	The subset of the immunogenicity analysis set who were seronegative at baseline.
Seropositive immunogenicity analysis set	The subset of the immunogenicity analysis set who were seropositive at baseline.
Seronegative safety analysis set	Subset of the Safety analysis set seronegative at baseline
Seropositive safety analysis set	Subset of the Safety analysis set seropositive at baseline

3.3 General Considerations

Descriptive analyses will support evaluation of safety, reactogenicity and immunogenicity.

Geometric mean titre ratios and differences in seroresponses with 95% confidence intervals will be presented to support selected comparisons of immunogenicity across groups of interest.

Immunogenicity against SARS-CoV-2 Wuhan-Hu-1 and B.1.351 strains will be characterized through the quantification of Spike-binding antibodies, pseudo-neutralization and, in a subset of participants, live neutralization. Exploratory analysis of immunogenicity against other strains and induction of other immune effectors including cell-mediated immunity will be conducted.

Categorical variables will be summarised using frequency and percentages, where the denominator for calculation is the underlying analysis set population, unless otherwise stated. A row denoted as “Missing” will be included in the count tabulations where necessary to account for missing values. Percentages will be reported with 2 decimal places. Summaries will be provided by treatment and visit when applicable.

Continuous variables will be summarised with descriptive statistics of number of participants with available data (N), mean, standard deviation (SD), median, minimum, and maximum. As appropriate, minima and maxima will be reported with the same precision as the raw values; medians and means will have one additional decimal place; standard deviation will have 2 additional decimal places. Summaries will be provided by treatment and visit when applicable.

Data will be displayed by treatment group in all listings, as needed. Participants will be uniquely identified in the listings by the combination of study number, study site number, and participant number.

All analyses will be performed using SAS®, Version 9.4 or higher (SAS Institute Inc., Cary, NC).

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

3.3.1 Baseline Definition

In general, the last non-missing measurement collected prior to the first dose is considered as baseline. Measurements collected on same day as dose are assumed to be prior to dose, unless timestamp shows the collection was after dose.

3.3.2 Serostatus Definition

The seronegative population of participants are defined as those which

1. had no history of laboratory-confirmed SARS-CoV-2 infection (i.e., no positive nucleic acid amplification test and no positive antibody test)
2. were assessed as negative for SARS-CoV-2 at screening following a lateral flow test to detect reactivity to the nucleoprotein.

Participants not meeting the above criteria are considered to be seropositive and included in the relevant analysis sets in Section 3.2.

3.3.3 Windowing Conventions

A windowing convention will be used to determine the analysis value for a given study visit for immunogenicity data analyses. The window definitions as following will be used for immunogenicity.

- A window of ± 7 days from the target day is applied to the following visits:
  - All participants: Days 15, 29.
  - Previously unvaccinated 4-week dosing interval: Days 43, 57.
  - Previously unvaccinated 12-week dosing interval: Days 99, 113.
- A window of ± 14 days from the target day is applied to the following visits: Day 85 (12-week dosing interval).
- A window of ± 28 days from the target day (relative to Dose 1 or Dose 2) is applied to the following visits:
  - Previously vaccinated: Day 180.
  - Previously unvaccinated 4-week dosing interval: Day 209.
  - Previously unvaccinated 12-week dosing interval: Day 265

One or more results for a particular immunogenicity endpoint may be obtained in the same visit window. In such an event, the result with the date closest to the expected visit date will be used in the analysis. In the event that two observations are equidistant from the expected visit date, the later observation will be used in the analysis.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Table 9 Visit Window for Immunogenicity

Participants	Dosing Period	Visit	Day Relative to Dose within the Dosing Period[a]	Visit Window (Study Day) Relative to the Dosing Period
Previously Vaccinated	Period 1 (Relative to Dose 1)	Baseline[b]	≤ 1	≤ 1
		Day 15	15	8-21
		Day 29	29	22-35
		Day 180	180	153-209
Previously Unvaccinated: 4-Week Dosing Interval	Period 1 (Relative to Dose 1)	Baseline[b]	≤ 1	≤ 1
		Day 15	15	8-21
		Day 29[c]	29	22-35
	Period 2 (Relative to Dose 2)	Day 43	14	8-21
		Day 57	28	22-35
		Day 209	180	153-209
Previously Unvaccinated: 12-Week Dosing Interval	Period 1 (Relative to Dose 1)	Baseline[b]	≤ 1	≤ 1
		Day 15	15	8-21
		Day 29	29	22-35
		Day 85[d]	85	71-98
	Period 2 (Relative to Dose 2)	Day 99	14	8-21
		Day 113	28	22-35
		Day 265	180	153-209

[a] For each dosing period, the administration of the study intervention is designated as Study Day 1. For analyses within a period, the study day value is incremented by 1 for each date following the vaccine administration. Dates prior to the vaccine administration are decremented by 1, with the date preceding the vaccine administration designated as Study Day -1.

[b] Where time is available, the time of the collection must be prior to the first dose of study intervention. Day 1 observations taken after the first dose are considered post-baseline values.

[c] For previously unvaccinated participants, Day 29 sample collection must occur before the 2nd vaccine administration to be included in analyses.

[d] For previously unvaccinated participants, Day 85 sample collection must occur before the 2nd vaccine administration to be included in analyses.

3.3.4 Handling of Unscheduled Visits

For overall analyses not based on any particular study visit, all data will be listed and/or analysed, including any repeated or unscheduled visits, unless otherwise specified.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

For the analyses based on particular study visit, please refer to windowing conventions for the visit determination and scheduled visit handling.

All collected data including unscheduled visits data will be listed in the listings.

3.3.5 Important Protocol Deviations

Important protocol deviations (IPDs) are a subset of protocol deviations (PDs) that may significantly impact the completeness, accuracy, and/or reliability of the study data or that may significantly affect a participant's rights, safety, or well-being.

The final list of IPDs will be documented prior to unblinding the study data and will include but may not be limited to the following deviations:

- Participant was randomised but did not meet inclusion criteria
- Participant was randomised but met exclusion criteria
- Participant developed discontinuation criteria but continued the IMP

IPDs will be summarised by the highest level deviation category and will be based on the full analysis set (FAS). IPDs or PDs related to the ongoing and emerging novel coronavirus (COVID-19) will be summarised for the FAS overall and by randomised treatment group. Participants with IPDs or PDs related to COVID-19 will be listed.

The immunogenicity analysis set includes all participants in the safety analysis set who have no protocol deviations or intercurrent events judged to have the potential to interfere with the generation or interpretation of an immune response. Analyses will exclude data from participants that occur after any of the above intercurrent events. Details of exclusionary protocol deviations will be defined in a separate PD plan.

3.4 Definitions of Subgroups

To explore the implications for safety and immunogenicity among different populations, the following subgroups will be used:

- Age at consent (18-64, 65 years and above)
- Gender (male, female)
- Comorbidity at baseline (at least one comorbidity vs no comorbidity), where comorbidity is BMI $\geq$ 30 kg/m^2 at baseline, cardiovascular disease, chronic lung disease or diabetes, except for historical control participants selected from D8110C00001 where the list of comorbidities is listed in Section 4.1.2.2.

The analyses by each subgroup will be performed for safety and immunogenicity endpoints, where appropriate.

3.5 Handling of Missing Data

As a general rule, missing data values will not be imputed unless otherwise specified.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

In the summary of prior/concomitant medications, missing or partially missing start/stop dates will be handled using the following imputation rules:

- The missing day of start date of a therapy will be set to the first day of the month that the event occurred.
- The missing day of end date of a therapy will be set to the last day of the month of the occurrence.
- If the start date of a therapy is missing both the day and month, the onset date will be set to January 1 of the year of onset.
- If the end date of a therapy is missing both the day and month, the date will be set to December 31 of the year of occurrence.
- If the start date of a therapy is null and the end date is not a complete date, the start date will be set to the date of the first study visit.
- If the start date of a therapy is null and the end date is a complete date and the end date is after the date of the first study visit, the start date will be set to the date of the first study visit. Otherwise, the start date will be set to the end date of the therapy.
- If the end date of a therapy is null and the start date is not a complete date, the end date will be set to the study end date.
- If the end date of a therapy is null and the start date is a complete date and the start date is prior to the study end date, the end date will be set to the study end date. Otherwise, the end date will be set to the start date of the therapy.

See Section 4.3.1.3 for handling of missing or partially missing start/stop dates for adverse events.

3.6 Reference Start Date and Study Day

Study Day will be calculated from the reference start date and will be used to show start/stop day of assessments and events. Reference start date is defined as the day of the first dose of study drug intervention ie, Day 1.

Study Day will be computed as follows:

- Study Day = (Date of event –Date of first dose of study drug + 1) for events on or after date of first dose
- Study Day = (Date of event –Date of first dose of study drug) for events before date of first dose
- Study Day (relative to second dose) = (Date of event –Date of second dose of study drug + 1)

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Day 1 observations made after the first dose are considered baseline values in cases where there is not an observation prior to administration of the booster. In all other cases, baseline will be the last observation prior to administration of the booster.

4 STATISTICAL ANALYSIS

4.1 Study Population

4.1.1 Participant Disposition and Completion Status

4.1.1.1 Definitions and Derivations

Disposition information can be directly obtained from the CRF.

4.1.1.2 Presentation

Disposition will be summarised by treatment group and overall using the all participants analysis set.

The disposition table of participants will include the number of participants with informed consent, screen failure, and the number and percentage of participants ongoing in study, completed study, discontinued early from treatment, discontinued early from study, and reasons of discontinuation. Only one reason for discontinuation will be recorded for each discontinued participant.

Disposition and screen failure information will be listed using the all participants analysis set.

4.1.2 Demographics and Baseline Characteristics

4.1.2.1 Definitions and Derivations

Most demography and baseline characteristic information can be directly obtained from the CRF. Age will be derived from the date of randomisation - date of birth, rounded down to the nearest integer. For participants in countries where date of birth is not recorded, the age as recorded in the electronic case report form will be used.

4.1.2.2 Presentation

Demography and baseline characteristics data will be summarised for the FAS and the immunogenicity analysis sets.

Demographics:

- Age (years) at consent
- Age group (18-64 vs ≥ 65 years)
- Gender
- Race (Asian, Black, White, Mixed, Other, Unknown)
- Country

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Baseline characteristics:

- Time since previous vaccination (for previously vaccinated individuals)
- BMI at baseline (< 30 vs ≥ 30 kg/m^2)
- Serostatus at baseline (Negative, Positive)
- Comorbidity at baseline – at least one of the following (Yes vs No)
  - Significant cardiovascular disease (Yes vs No)
  - Chronic lung disease (Yes vs No)
  - Diabetes (Yes vs No)

The following will be summarised for the historical control participants with a primary vaccination of AZD1222 with no booster and for the pooled set of participants in the seronegative immunogenicity analysis set previously vaccinated with AZD1222 and receiving a booster, and the pooled set of participants in the seronegative immunogenicity analysis set previously vaccinated with a mRNA vaccine and receiving a booster:

- Age (years) at consent
- Age group (18-64 vs 65 and older)
- Sex
- Comorbidity at baseline (At Least One vs None)
- BMI at baseline ≥30 kg/m2 (Yes vs No)
- BMI at baseline (kg/m2)

In the D8110C00001 study, historical control participants were defined as having at least one comorbidity at baseline if they had history of any of the following:

- Chronic kidney disease
- Chronic obstructive pulmonary disease (COPD), like emphysema
- Lower immune health because of a solid organ transplant
- Obesity – those with a BMI greater than 30
- Serious heart condition like heart failure and coronary artery disease
- Sickle cell disease
- Type 2 diabetes
- Asthma
- Dementia
- Cerebrovascular diseases, such as stroke
- Cystic fibrosis
- High blood pressure
- Liver disease

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

- Scarring in the lungs (pulmonary fibrosis)
- Type I diabetes
- Thalassemia (a blood disorder)
- Smoking

4.1.3 Prior and Concomitant Medications

4.1.3.1 Definitions and Derivations

A medication will be regarded as prior if it started prior to the date of randomisation and was stopped on or before the date of randomisation (medication stop date ≤ date of randomisation).

A medication will be regarded as concomitant if the start date is on or after the date of randomisation, or if it started prior to the date of randomisation and was ongoing after the date of randomisation. Medications with start date after the on-treatment period will not be considered as concomitant.

Partial date conventions are detailed in Appendix 1. Partial Date Conventions.

4.1.3.2 Presentation

The number and percentage of participants who take prior medications, those who take allowed concomitant medications and those who take medications that are considered intercurrent events during the study, will be presented by treatment group and 'Total'. Concomitant medications will be classified according to the WHO-Drug Dictionary. The summary tables will present data by generic term within ATC code.

4.1.4 Exposure

4.1.4.1 Definitions and Derivations

Dosing interval for two dose participants is calculated in weeks as:

Dosing interval =(second dose date of study drug - first dose date of study drug)/7

4.1.4.2 Presentation

The exposure information including number of dose(s) and all available dose intervals for two doses and further categorized dose schedule will be summarised by treatment for the safety analysis set.

A listing of exposure will be provided at the time of the primary analysis.

4.2 Immunogenicity Analyses

4.2.1 Previously unvaccinated cohort receiving 2-dose primary vaccination

Please refer to the Naïve-SAP.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

4.2.2 Previously vaccinated cohort receiving 1-dose booster vaccination following two doses of AZD1222.

Please refer to the V1222-SAP.

4.2.3 Previously vaccinated cohort receiving 1-dose booster vaccination following two doses of a mRNA vaccine.

Please refer to the VmRNA-SAP.

4.3 Safety Analyses

The domain safety covers exposure, adverse events, clinical laboratory, and vital signs.

Tables are provided for the safety analysis set; listings are provided for the all participants or the safety analysis set depending on the availability of data. Safety summaries described below will be performed by treatment group.

4.3.1 Adverse Events

4.3.1.1 Definitions and Derivations

Adverse event severity will be graded according to a revised toxicity grading scale from the US FDA guidance (FDA 2007) and coded using the most recent version of the Medical Dictionary for Regulatory Activities.

The safety of AZD2816 and AZD1222 will assessed by:

- Incidence of local and systemic solicited adverse events (AEs) for 7 days post dose
- Incidence of unsolicited AEs, serious adverse events (SAEs), medically attended adverse events (MAAEs), and adverse events of special interest (AESIs) for 28 days post dose
- Incidence of MAAEs, SAEs, and AESIs from post first dose through 6 months post last dose

Solicited and Unsolicited AEs

Solicited AEs are local or systemic predefined events for assessment of reactogenicity. Solicited AEs will be collected in a e-diary for 7 days following administration of each dose and will be assessed separately from the (unsolicited) AEs collected during the study. If a solicited AE is not resolved within the e-diary reporting period, the event will also be reported as an unsolicited adverse event in the eCRF, with actual start and stop dates.

Solicited AEs should not be reported as unsolicited AEs unless they fulfil the criteria for SAEs or medically-attended AEs. All other AEs are considered to be unsolicited AEs.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Table 10 Predefined Solicited Adverse Events for Reactogenicity Assessment

Local	Systemic
Pain at the site of the injection	Fever (> 100 °F/37.8 °C)
Redness/erythema at the site of the injection	Chills
Tenderness at the site of the injection	Muscle pains
Induration/swelling at the site of the injection	Fatigue (physical or mental tiredness/exhaustion)
-	Headache
-	Malaise (general feeling of discomfort or uneasiness)
-	Nausea
-	Vomiting

AEs and SAEs

AEs will be recorded for 28 days after each dose of study intervention. SAEs are those events recorded as "Serious" on the AE page of the eCRF. SAEs will be recorded from the time of signing the informed consent form through the last participant contact. The definition of AEs and SAEs can be found in Protocol Appendix B.

Listings of AEs and SAEs will be provided. AEs and SAEs prior to the dose of IMP will only be presented in the listings. For SAEs with partial dates, if the known part of the date indicates that SAE stopped before the dose of IMP, it will be considered as SAE prior to the dose of IMP. Otherwise, it will be considered as SAE post dose of IMP.

MAAEs

Medically-attended AEs are defined as AEs leading to medically-attended visits that were not routine visits for physical examination or vaccination, such as an emergency room visit, or an otherwise unscheduled visit to or from medical personnel (medical doctor) for any reason. AEs, including abnormal vital signs, identified on a routine study visit or during the scheduled illness visits will not be considered medically-attended AEs.

AESIs

AESIs are events of scientific and medical interest, specific to the further understanding of the IMP safety profile and require close monitoring and rapid communication by the investigators to the Sponsor. An AESI can be serious or non-serious. All AESIs will be recorded in the eCRF. AESIs for this study are listed in Appendix E of the CSP.

COVID-19

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

COVID-19 is defined as SARS-CoV 2-RT-PCR positive symptomatic illness. At all clinic visits following the initial vaccination, participants will be asked if they have had a diagnosis of COVID-19 since their last clinic visit. Medical records will be obtained for confirmation of a participant-reported diagnosis of COVID-19. Qualifying symptoms are fever, shortness of breath, difficulty breathing, chills, cough, fatigue, muscle/body aches, headache, new loss of taste or smell, sore throat, congestion, runny nose, nausea, vomiting, or diarrhoea. Events will be reported as AEs/SAEs. If a participant presents at clinic visit with COVID symptoms, diagnosis will be confirmed using RT-PCR.

Deaths

If any participants die during the study as recorded on the "Death Details" page of the eCRF, the number and percentage of participants with death related to COVID-19 and those with other deaths will be summarised by actual treatment group based on the safety analysis set.

Severity

Summary of AEs and SAEs post the dose of IMP will be broken down further by maximum severity and relationship to study intervention. Severity will be classified as mild, moderate, severe, life-threatening, and fatal. Severity for AEs will be collected on "Adverse Events" form of eCRF. Should a participant experience multiple events within a system organ class (SOC) or preferred term (PT), only the participant's worst severity grade will be counted for that SOC or PT.

Relationship to IMP

Relationship to IMP/other medication/study procedure, as indicated by the Investigator, will be classified as not related or related. Should a participant experience multiple events within a SOC or PT, the participant will be counted as related for that SOC or PT if one of those is related.

Exposure adjusted rate

Exposure adjusted rate is calculated as number of participants with AEs in categories divided by total participant-year exposure to investigational study intervention. Participant years is determined by summing the total number of follow-up days of each participant, and then dividing by 365.25. The exposure period is calculated from time of dose to end of study for safety endpoints measured over 6 months post-dose, or from time of dose to 28 days post dose for safety endpoints measured over 28 days post-dose.

4.3.1.2 Presentation

AEs will be presented for actual treatment for the safety analysis set in categories as below.

- AEs
- SAEs

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

- SAEs with outcome death
- AEs leading to IMP discontinuation
- AEs leading to study discontinuation
- Related AEs by investigator's causality
- MAAEs
- AESIs

Summaries will include the number and percentage of participants reporting at least one event, number of events, and exposure adjusted rates, where appropriate.

An overview table of AEs will be presented for actual treatment, including the number and percentage of participants for each category.

Moreover, each category will be presented by SOC and PT. Should a participant experience multiple events within a category, the participant will be counted only once for that category.

The incidence of local and systemic solicited AEs following each vaccination will be summarised by day and overall for 7 days following each vaccination. The incidence of unsolicited AEs, SAEs, MAAEs, and AESIs will be summarised for 28 days following each vaccination. The incidence of MAAEs, SAEs, and AESIs will be summarised following the first vaccination and throughout the study.

A summary of the most common AEs will be presented by PT. Additionally, a summary of non-serious AEs occurring in >5% of participants in any treatment group will be presented by PT. AEs and SAEs causing discontinuation of the study treatment and SAEs causing discontinuation from the study will also be summarised by PT.

AEs and SAEs will be summarised by preferred term and maximum intensity. Related AEs and SAEs will also be summarised by preferred term. If a participant reports multiple occurrences of the same AE within the same study period, the maximum intensity will be taken as the highest recorded maximum intensity (the order being mild, moderate, and severe).

The incidence of COVID-19 AEs will be summarised from the first dose to the study end.

AEs that occur after unblinding due to vaccination (defined as the earliest of the date of early unblinding and the date of receiving an unblinded non-study COVID-19 vaccination) will be presented and tabulated separately.

4.3.1.3 Handling of Missing Data

In the summary of AEs by timing relative to each vaccination, if the AE onset date is completely missing, then the AE will be included in the summary. If the AE onset date is partially missing, partial AE dates will be handled using the following imputation rules:

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

- Partial AE start dates where only the year is known:
  - If the year is the same as the year of dosing:
    - Assume (first dose date + 1 day) if when AE occurred relative to dosing is not available
    - Assume dose date otherwise
  - If the year is not the same as the year of dosing, assume January 1 for start date
- Partial AE stop dates where only the year is known: assume December 31 for stop date. If imputed date is greater than the cut-off date of data, use cut-off date instead.
- Partial AE start dates where only the month and year are known:
  - If the year and the month are the same as the year and the month of dosing:
    - Assume (dose date + 1 day) if when AE occurred relative to dosing is not available
    - Assume dose date otherwise
  - If the year and the month is not the same as the year and the month of dosing: assume the first of the month for start date.
- Partial AE stop dates where only the month and year are known: assume the end of the month for stop date

4.3.2 Clinical Laboratory, Blood Sample and Urinalysis

4.3.2.1 Definitions and Derivations

Hematology and serum clinical chemistry will be performed as per the schedule of events (refer to the CSP for schedules and lists of parameters). A urine pregnancy test will be performed at screening.

Quantitative laboratory parameters reported as "< X", i.e. below the lower limit of quantification or "> X", i.e. above the upper limit of quantification, will be converted to X for the purpose of quantitative summaries, but will be presented as recorded, i.e. as "< X" or "> X" in the listings.

Quantitative laboratory parameters will be compared with the relevant central laboratory normal ranges in SI units and categorized as:

- Low: Below the lower limit of the laboratory normal range.
- Normal: Within the laboratory normal range (upper and lower limit included).
- High: Above the upper limit of the laboratory normal range.

Quantitative laboratory parameters with available FDA toxicity grades will be categorized as follows where higher grades representing a more severe toxicity (refer to Appendix 2. FDA Laboratory Abnormality Severity Grade Criteria for a partial list of parameter toxicity grade criteria):

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

- Grade 1 (i.e., mild);
- Grade 2 (i.e., moderate);
- Grade 3 (i.e., severe);
- Grade 4 (i.e., life-threatening);

Non-missing laboratory parameter results not meeting any of the 4 grades defined in the FDA toxicity grading system will be categorized as 'No Event' for the purpose of the shift from baseline summaries.

4.3.2.2 Presentations

The change from baseline for laboratory measures will be summarised at time points as detailed in the schedule of assessment. The following summaries will be provided by actual treatment group for laboratory parameters:

- Observed and change from baseline in Standard International (SI) units by visit (for quantitative parameters).
- Number and percentage of participants in each laboratory parameter category by visit (for categorical parameters).
- Maximum post-baseline ALT/AST observed value categorized as < 3 x upper limit of normal (ULN), ≥ 3 to < 5 x ULN, ≥ 5 to < 10 x ULN or ≥ 10 ULN by maximum post-baseline total bilirubin (TBL) observed value categorized as < 2 x ULN or ≥ 2 x ULN.
- A listing of participants with at least one observed value in ALT value ≥ 3 x ULN, AST value ≥ 3 x ULN or TBL value ≥ 2 x ULN will be provided.
- Shift from baseline to the worst post-baseline observed value according to the FDA toxicity grades (for quantitative parameters with available FDA toxicity grades.
- Shifts from baseline to the maximum/minimum post-baseline observed value according to normal range criteria.
- Listing of participants with at least one abnormal laboratory observed value outside the normal range criteria.

4.3.3 Vital Signs

4.3.3.1 Definitions and Derivations

Vital signs will be performed at time points specified in Protocol Section 1.3, including the following parameters:

- Systolic blood pressure (mmHg)
- Diastolic blood pressure (mmHg)

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

- Heart rate (beats per minute)
- Body temperature (C)
- Oxygen saturation (%)

4.3.3.2 Presentations

For severity grades of abnormal Vital Signs refer to Appendix 3. Clinical Abnormalities: Vital Signs.

The following summaries will be provided by actual treatment group for each vital sign parameter:

- Observed and change from baseline by visit
- Number and percentages of participants with at least one abnormal post-baseline observed value (refer to Appendix 3. Clinical Abnormalities: Vital Signs)

4.3.4 Other Safety Assessments

4.3.4.1 Physical Examination

Physical examinations (completed and targeted) will be conducted as per the schedule of events (refer to protocol Section 1.3). Clinically significant abnormal findings at screening will be recorded in the Medical History, while clinically significant abnormal findings following vaccination will be recorded as AEs.

5 INTERIM ANALYSIS

Details of the initial and third interim analyses are provided in the Interim Analysis Charter.

Details of the second analysis are provided in the V1222-Sap and the VmRNA-SAP.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

6 REFERENCES

FDA. (Food and Drug Administration). Guidance for Industry. Toxicity grading scale for healthy adult and adolescent volunteers enrolled in preventive vaccine clinical trials. https://www.fda.gov/media/73679/download. Published 2007. Accessed 20 June 2020.

Little, R. J. A. and Rubin, D. B. Statistical Analysis with Missing Data, 2nd Edition, Hoboken, NJ: John Wiley & Sons 2002; 257.

NIH. (National Institutes of Health) National Institute of Allergy and Infectious Diseases, Division of AIDS. Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1 [July 2017]. https://rsc.niaid.nih.gov/sites/default/files/daidsgradingcorrectedv21.pdf. Published 2017.

CHMP (Committee for Human Medicinal Products). Reflection paper on the regulatory requirements for vaccines intended to provide protection against variant strain(s) of SARS-CoV-2. https://www.ema.europa.eu/en/documents/scientific-guideline/reflection-paper-regulatory-requirements-vaccines-intended-provide-protection-against-variant_en.pdf. Published 2021.

Hing, J.P., Woolfrey, S.G., Greenslade, D. et al. Analysis of Toxicokinetic Data Using NONMEM: Impact of Quantification Limit and Replacement Strategies for Censored Data. J Pharmacokinet Pharmacodyn 28, 465–479 (2001). https://doi.org/10.1023/A:1012247131190

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

7 APPENDIX

Appendix 1. Partial Date Conventions

Algorithm for Prior / Concomitant Medications

START DATE	STOP DATE	ACTION
Known	Known or ongoing	If medication stop date < date of dose of IMP, assign as prior; If medication start date < date of dose of IMP and medication stop date ≥ date of dose of IMP, assign as concomitant; If date of dose of IMP ≤ medication start date, assign as concomitant.
	Partial	If known components of medication stop date show that medication stopped before date of dose of IMP, assign as prior; If medication start date < date of dose of IMP and (known components of medication stop date show that medication stopped on or after date of dose of IMP), assign as concomitant; If date of dose of IMP ≤ medication start date, assign as concomitant.
	Missing, not ongoing	If medication stop date is missing, then it can never be assigned as prior only; If medication start date < date of dose of IMP, assign as concomitant; If date of dose of IMP ≤ medication start date, assign as concomitant.
Partial	Known or ongoing	If medication stop date < date of dose of IMP, assign as prior; If (known components of medication start date show that medication started before date of dose of IMP) and (medication stop date ≥ date of dose of IMP), assign as concomitant; If known components of medication start date show that medication started on or after date of dose of IMP, assign as concomitant.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

START DATE	STOP DATE	ACTION
	Partial	If known components of medication stop date show that medication stopped before date of dose of IMP, assign as prior; If (known components of medication start date show that medication started before date of dose of IMP) and (known components of medication stop date show that medication stopped on or after date of dose of IMP), assign as concomitant; If known components of medication start date show that medication started on or after date of dose of IMP, assign as concomitant.
	Missing, not ongoing	Cannot be assigned as prior only; If known components of medication start date show that medication started before study drug start date, assign as concomitant; If known components of medication start date show that medication started on or after date of dose of IMP, assign as concomitant.
Missing	Known or ongoing	If medication stop date < date of dose of IMP, assign as prior; If medication stop date ≥ date of dose of IMP, assign as concomitant.
	Partial	If known components of medication stop date show that medication stopped before date of dose of IMP, assign as prior; If known components of medication stop date show that medication stopped on or after date of dose of IMP, assign as concomitant.
	Missing, not ongoing	Assign as concomitant.

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Appendix 2. FDA Laboratory Abnormality Severity Grade Criteria

Variable	Unit	Grade 1	Grade 2	Grade 3	Grade 4
Haemoglobin Absolute Decreased (male)	g/L	125-135	105-124	85-104	<85
Haemoglobin Absolute Decreased (female)	g/L	110-120	95-109	80-94	<80
Haemoglobin Decrease from Baseline	g/L	1-15	16-20	21-50	>50
White Blood Cells-Elevated	cells x 10^9/L	10.8-15	>15-20	>20-25	>25
White Blood Cells-Decreased	cells x 10^9/L	2.5-3.5	1.5-2.49	1.0-1.49	<1.0
Platelets-Decreased	cells x 10^9/L	125-140	100-124	25-99	<25
Neutrophils-Decreased	cells x 10^9/L	1.5-2.00	1.0-1.49	0.5- 0.99	<0.50
Lymphocytes-Decreased	cells x 10^9/L	0.750-1.000	0.500-0.749	0.250-0.499	<0.250
Eosinophils-Elevated	cells x 10^9/L	0.650-1.500	1.501-5.000	>5.000	Hypereosinophilic
Creatinine-Elevated (converted from mg/dL)	mg/dL (µmol/L)	1.5-1.7 (133-154)	1.8-2.0 (155-181)	2.1-2.5 (182-221)	>2.5 (>221) or requires dialysis
Bilirubin-Elevated (with normal ALT/ALP)	µmol/L	1.1-1.5 × ULN	1.6-2.0 x ULN	2.0-3.0 × ULN	>3.0 × ULN
Bilirubin-Elevated (with abnormal ALT/ALP)	µmol/L	1.1-1.25 × ULN	1.26-1.5 × ULN	1.51-1.75 × ULN	>1.75 × ULN

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Alanine Transaminase-Elevated	U/L	1.1-2.5 x ULN	2.6-5 × ULN	5.1-10 × ULN	>10 × ULN
Alkaline Phosphate-Elevated	µmol/L	1.1-2.0 × ULN	2.1-3.0 × ULN	3.1-10 × ULN	>10 × ULN
Aspartate Transaminase-Elevated	U/L	1.1-2.5 x ULN	2.6-5 × ULN	5.1-10 × ULN	>10 × ULN
Prothrombin Time	seconds	1.0-1.10 x ULN	1.11-1.20 x ULN	1.21-1.25 x ULN	>1.25 ULN
Activated Partial Thromboplastin Time	seconds	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)

DocuSign Envelope ID: 7B0DC020-4183-47C3-967E-EF1F118FF724

Appendix 3. Clinical Abnormalities: Vital Signs

Vital Signs	Vital Signs Grade			
	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Fever (°C) (°F)	37.9-38.4 100.1-101.1	38.5-38.9 101.2-102.0	39.0-40 102.1-104	> 40 > 104
Tachycardia (beats/minute)	101-115	116- 130	> 130	ER visit or hospitalization for arrhythmia
Bradycardia (beats/minute)	50-54	45-49	< 45	ER visit or hospitalization for arrhythmia
Hypertension; systolic (mm Hg)	141-150	151-155	> 155	ER visit or hospitalization for malignant hypertension
Hypertension; diastolic (mm Hg)	91-95	96-100	> 100	ER visit or hospitalization for malignant hypertension
Hypotension; systolic (mm Hg)	85-89	80-84	< 80	ER visit or hospitalization for hypotensive shock

ER = emergency room.

Due to technical issues, PPD (PPD) (ClinChoice) was not able to sign in ANGEL/CARA, but is signing instead in DocuSign.

DocuSigned by PPD
PPD
I am the author of this document
23 September 2022 | 18:42 GMT
8C9818B7DD7449E0A7174A550A2608ED

DocuSign

Certificate Of Completion

Envelope Id: 7B0DC020418347C3967EEF1F118FF724
Status: Completed
Subject: Please DocuSign: d7220c00001-master-sap-ed-5.0.docx
Source Envelope:
Document Pages: 45
Signatures: 1
Envelope Originator:
Certificate Pages: 3
Initials: 0
PPD
AutoNav: Enabled
PPD
EnvelopeId Stamping: Enabled
PPD
Time Zone: (UTC+05:30) Chennai, Kolkata, Mumbai, New Delhi
PPD
IP Address: 24.30.87.232

Record Tracking

Status: Original
9/23/2022 11:25:42 PM
Holder: PPD
PPD
Location: DocuSign

Signer Events	Signature	Timestamp
PPD PPD Security Level: Email, Account Authentication (Required), Logged in	PPD Signature Adoption: Pre-selected Style Signature ID: 8C9818B7-DD74-49E0-A717-4A550A2608ED Using IP Address: 70.63.230.6 With Signing Authentication via DocuSign password With Signing Reasons (on each tab): I am the author of this document	Sent: 9/23/2022 11:27:36 PM Viewed: 9/24/2022 12:12:00 AM Signed: 9/24/2022 12:12:21 AM

Electronic Record and Signature Disclosure:
Accepted: 9/24/2022 12:12:00 AM
ID: 29b7a887-3279-4500-84b4-b90413c9b934

In Person Signer Events	Signature	Timestamp

Editor Delivery Events	Status	Timestamp

Agent Delivery Events	Status	Timestamp

Intermediary Delivery Events	Status	Timestamp

Certified Delivery Events	Status	Timestamp

Carbon Copy Events	Status	Timestamp

Witness Events	Signature	Timestamp

Notary Events	Signature	Timestamp

Envelope Summary Events	Status	Timestamps
Envelope Sent	Hashed/Encrypted	9/23/2022 11:27:36 PM
Certified Delivered	Security Checked	9/24/2022 12:12:00 AM
Signing Complete	Security Checked	9/24/2022 12:12:21 AM
Completed	Security Checked	9/24/2022 12:12:21 AM

Payment Events	Status	Timestamps

Electronic Record and Signature Disclosure

I hereby consent to that AstraZeneca Worldwide https://www.astrazeneca.com may disclose personal information such as; full name, email address, and any other information you may supply on the electronic form to AstraZeneca affiliates and third party service providers throughout the world in relation to the handling and administration of the Electronic Signature Service solution. This consent relates to any electronic records or signatures associated with the electronic contract.

AstraZeneca and the third party administering this service store and process personal information that AstraZeneca collects from you for the purposes of operating the Electronic Signature Service solution.

This also applies after termination of the Agreement. Processing of your personal information will be done in accordance with applicable law.

You may request access to your personal data and withdraw agreement to this processing at any time by contacting us in writing at docusign-support@astrazeneca.com.

Personal details and electronic signatures of signatories contained in contracts cannot be removed once the contract has been executed and will remain part of such contracts until these are destroyed in accordance with applicable law and AstraZeneca internal data retention policies.

SIGNATURE PAGE

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature

Document Name: d7220c00001-master-sap-ed-5		
Document Title:	D7220C00001 Master Statistical Analysis Plan Edition 5	
Document ID:	Doc ID-004787735	
Version Label:	2.0 CURRENT LATEST APPROVED	
Server Date **(dd-MMM-yyyy HH:mm 'UTC'Z)**	**Signed by**	**Meaning of Signature**
23-Sep-2022 20:51 UTC	PPD	Author Approval
23-Sep-2022 23:26 UTC	PPD	Content Approval

Notes: (1) Document details as stored in ANGEL, an AstraZeneca document management system.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

STATISTICAL ANALYSIS PLAN	
Study Code	D7220C00001
Edition Number	2.0
Date	23-Sep-2022

A Phase II/III Partially Double-Blinded, Randomised, Multinational, Active-Controlled Study in Both Previously Vaccinated and Unvaccinated Adults to Determine the Safety and Immunogenicity of AZD2816, a Vaccine for the Prevention of COVID-19 Caused by Variant Strains of SARS-CoV-2

(Previously Unvaccinated Cohort Sub-SAP)

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

TABLE OF CONTENTS

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

LIST OF TABLES

LIST OF FIGURES

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

LIST OF ABBREVIATIONS

Abbreviation or Specialized Term	Definition
AE	Adverse event
AESI	Adverse event of special interest
ANCOVA	Analysis of covariance
BMI	Body mass index
CI	Confidence Interval
CMI	Cell-mediated immune
COVID-19	Coronavirus 2019
CRF	Case report form
CSP	Clinical study protocol
GMFR	Geometric mean fold rises
GMT	Geometric mean titre
IMP	Investigational medicinal product
LLOQ	Lower limit of quantification
MAAE	Medically attended adverse event
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus 2
SD	Standard deviation
ULOQ	Upper limit of quantification

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

AMENDMENT HISTORY

CATEGORY* Change refers to:	Date	Description of change	In line with CSP?	Rationale
N/A	01 February, 2022	Initial approved SAP	N/A	N/A
Statistical analysis method for the primary or secondary endpoints	23 September, 2022	Updated random effects of pairwise comparisons	No	Further clarification of previous model specification
Other	23 September, 2022	Changed value for adjustment of continuous variables to be standardized across all subjects for sensitivity analyses Deleted irrelevant reference, and added 2 new references	No	To be provide a fair comparison between treatment arms for separate models More complete documentation of methods

*Pre-specified categories are:

Primary or secondary endpoints; Statistical analysis method for the primary or secondary endpoints; Derivation of primary or secondary endpoints; Multiple Testing Procedure; Data presentations; Other

N/A = Not applicable

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

1 INTRODUCTION

The purpose of this document is to give details for the statistical analysis of study D7220C00001 supporting the clinical study report for previously unvaccinated participants. The reader is referred to the Clinical Study Protocol (CSP), amendment 3 dated 11 October 2021 as well as the most recent versions of the Protocol Deviation Management Plan and the Case Report Form (CRF) for details of study conduct and data collection, and the master Statistical Analysis Plan (master-SAP) where referenced for details of the analysis principles applicable to all sub Statistical Analysis Plans (SAPs).

The term IMP (investigational medicinal product) is used throughout this SAP to include both treatment groups (AZD2816 and AZD1222).

1.1 Objectives and Endpoints

The primary safety objective for the cohort of previously unvaccinated participants receiving a 2-dose dose primary vaccination is to characterize the safety and tolerability of a 2-dose primary vaccination with AZD2816 with a 4-week dosing interval in previously unvaccinated seronegative participants.

The primary and key secondary immunogenicity objectives for this cohort are as follows:

Co-Primary:

- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval.
- To determine if seroresponse against the B.1.351 variant elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to seroresponse against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval**.**

Key secondary:

- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval.
- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by a 2-dose heterologous AZD1222 + AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval.

The above primary and the key secondary immunogenicity objectives will be supported by other secondary immunogenicity objectives (see below) for which are not included in the testing hierarchy.

Table 1 further describes the objectives and endpoints for this cohort of participants, including estimands for the immunogenicity objectives.

Table 1 Study Objectives and Endpoints for Previously Unvaccinated Participants Receiving a 2-Dose Primary Vaccination

Safety Objectives	
Objectives	**Endpoints**
- Primary	
To characterize the safety and tolerability of a 2-dose primary vaccination with AZD2816 with a 4-week dosing interval in previously unvaccinated seronegative participants	• Incidence of local and systemic solicited AEs for 7 days post-dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs, for 28 days post-dose • The change from baseline for safety laboratory measures for 28 days post-dose
- Secondary	
To characterize the safety and tolerability of a heterologous primary vaccination with 1 dose of AZD1222 followed by 1 dose of AZD2816 administered with a 4-week dosing interval in previously unvaccinated seronegative participants	• Incidence of local and systemic solicited AEs for 7 days post-dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs for 28 days post-dose
To characterize the safety and tolerability of a 2-dose primary vaccination with AZD2816 with a 12-week dosing interval in previously unvaccinated seronegative participants	• Incidence of local and systemic solicited AEs for 7 days post-dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs for 28 days post-dose
To characterize the extended safety of a 2-dose primary vaccination with AZD2816 with a 4-week dosing interval in previously unvaccinated seronegative participants	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
To characterize the extended safety and tolerability of a heterologous primary vaccination with 1 dose of AZD1222 followed by 1 dose of AZD2816 administered with a 4-week dosing interval in previously unvaccinated seronegative participants	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
To characterize the extended safety of a 2-dose primary vaccination with AZD2816 with a 12-week dosing	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

interval in previously unvaccinated seronegative participants		
Immunogenicity objectives		
To determine if the pseudoneutralizing antibody GMT response elicited by a 2-dose AZD2816 primary vaccination with a 4-week dosing interval is non-inferior to the response elicited by a 2-dose AZD1222 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD2816 vaccination	AZD1222 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective Level:	Serotype comparison:	
Primary	B.1.351	Wuhan-Hu-1
Key Secondary 2.1	B.1.351	B.1.351
Key Secondary 2.3	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD2816 vaccination/AZD1222 vaccination	
To determine if the seroresponse rate elicited by a 2-dose AZD2816 primary vaccination with a 4-week dosing interval is non-inferior to the seroresponse rate elicited by a 2-dose AZD1222 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD2816 vaccination	AZD1222 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective Level:	Serotype comparison:	
Co-primary	B.1.351	Wuhan-Hu-1
Other Secondary	B.1.351	B.1.351
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 vaccination - AZD1222 vaccination	
To determine if the neutralizing antibody GMT response elicited by a 2-dose AZD1222+AZD2816 heterologous primary vaccination with a 4-week dosing interval is non-inferior to the response elicited by a 2-dose AZD1222 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD1222+AZD2816 vaccination	AZD1222 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective Level:	Serotype comparison:	
Key Secondary 2.2	B.1.351	Wuhan-Hu-1
Other Secondary	B.1.351	B.1.351
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD1222+AZD2816 vaccination/AZD1222 vaccination	
To determine if the seroresponse rate elicited by a 2-dose AZD1222+AZD2816 heterologous primary vaccination with a 4-week dosing interval is non-inferior to the seroresponse rate elicited by a 2-dose AZD1222 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD1222+AZD2816 vaccination	AZD1222 vaccination

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective Level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Other Secondary	B.1.351	B.1.351
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD1222+AZD2816 vaccination - AZD1222 vaccination	
To determine if the neutralizing antibody GMT response against the B.1.351 variant is non-inferior to the response against the original Wuhan-Hu-1 strain following a 2-dose AZD2816 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD2816 vaccination	AZD2816 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following a 2-dose AZD2816 primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD2816 vaccination	AZD2816 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) against B.1.351 versus Wuhan-Hu-1	
To determine if the neutralizing antibody GMT response against the B.1.351 variant is non-inferior to the response against the original Wuhan-Hu-1 strain following a 2-dose AZD1222+AZD2816 heterologous primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD1222+AZD2816 vaccination	AZD1222+AZD2816 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks
Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following a 2-dose AZD1222+AZD2816 heterologous primary vaccination with a 4-week dosing interval Estimand:		
Treatment	AZD1222+AZD2816 vaccination	AZD1222+AZD2816 vaccination
Population	Seronegative participants with no prior COVID-19 vaccination	Seronegative participants with no prior COVID-19 vaccination
Dosing interval	4 weeks	4 weeks

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Timepoint	28 days after second vaccination dose	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) against B.1.351 versus Wuhan-Hu-1	
To also determine non-inferiority of the s-protein binding antibody response for the above comparisons as other secondary objectives		
To also determine the neutralizing antibody GMT responses 28 days after first vaccination dose in the above primary and key secondary objectives		
To explore anti-vector responses to the ChAdOx-1 adenovirus vector following a 2-dose homologous or heterologous primary vaccination with AZD2816 and/or AZD1222 in sub-groups of seronegative and seropositive participants	• GMT of ChAdOx1 neutralizing antibody titres • Seroresponse rate of ChAdOx1 neutralizing antibody titres Pairwise correlations between anti-S, pseudo-neutralization, and ChAdOx1 neutralizing antibody titres, 28 days after both Dose 1 and Dose 2	
Exploratory objectives		

CCI

[redacted]

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

CCI

MAAEs: medically attended adverse events; SAEs: serious adverse events; AESIs: adverse events of special interest
Seroresponse: An at least 4-fold increase in geometric mean titre from baseline.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

1.2 Study Design

Please refer to the master-SAP.

1.3 Sample Size

Please refer to the master-SAP.

2 CHANGES TO PROTOCOL PLANNED ANALYSES

By request of the Committee for Medicinal Products for Human Use (CHMP), separate SAPs have been created to analyze participants previously vaccinated with AZD1222, participants previously vaccinated with a mRNA vaccination, and previously unvaccinated participants. As a result, this SAP only specifies endpoints relevant to previously unvaccinated participants.

For further details to changes to the protocol planned analysis following feedback from the EMA and Medicines and Healthcare products Regulatory Agency (MHRA) please refer to the master-SAP.

3 DATA ANALYSIS CONSIDERATIONS

Please refer to the master-SAP.

4 STATISTICAL ANALYSIS

4.1 Study Population

Please refer to the master-SAP.

4.2 Immunogenicity Analyses

4.2.1 Humoral immune response following vaccination

For previously unvaccinated participants randomised to a 4-week dosing interval, testing for antibody responses will be performed at baseline, day 15, day 29, day 43, day 57, and day 209.

For previously unvaccinated participants randomised to a 12-week dosing interval, testing for antibody responses will be performed at baseline, day 15, day 29, day 85, day 99, day 113, and day 265.

The immunogenicity endpoints of interest in this study are:

- Geometric mean antibody titre
- Seroresponse, defined as ≥ 4-fold increase in the geometric mean antibody titre from baseline

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Both the geometric mean antibody titre and seroresponse of participants will be summarised descriptively by strain, treatment received, and timepoint for the immunogenicity analysis set. Scatter plots with box plot overlay for geometric mean antibody titre will be presented by strain, treatment received, and timepoint for the immunogenicity analysis set. Analyses will be performed for each of the following analysis sets:

- Immunogenicity analysis set
- Seronegative immunogenicity analysis set
- Seropositive immunogenicity analysis set

The following intercurrent events could impact the antibody levels achieved:

- missing the second vaccination (for the unvaccinated population)
- receiving of immune-modifying drugs or vaccines
- SARS-CoV-2 infection during the study (COVID-19, SARS-CoV-2 test positive, asymptomatic COVID-19, COVID-19 pneumonia)

All immunogenicity descriptions and comparisons will use the principal stratum strategy, ie, all analyses will exclude data from participants that occur after any of the above intercurrent events.

The following antibody titre measurements from serum samples will be analysed:

- Spike-specific IgG response to SARS-CoV-2 by multiplexed immunoassay (S antibody)
- Antibody neutralisation using a lentivirus-based pseudovirus particle expressing the SARS-CoV-2 spike protein [neutralizing antibody (pseudoneutralization)]
- ChAdOx1 neutralizing antibody titres

CCI

Antibody responses will be tested against the following strains:

- Wuhan-Hu-1
- B.1.351 (beta)
- B.1.617.2 (delta)

Other variants of concern or interest may be analysed as exploratory analyses.

All assay results will be presented in units in which the assay is performed. Should an international standard be released, then immunogenicity analyses may be repeated using the international standards.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

4.2.2 CCI

CCI

[redacted]

4.2.3 Statistical Hypothesis

The overall hypothesis for previously unvaccinated participants is that 28 days after the 2nd dose with a 4-week dosing interval, AZD2816 will be non-inferior to AZD1222 in terms of immunogenicity. This will be concluded only if both the co-primary endpoints (GMT ratio and difference in seroresponse) are met.

All non-inferiority comparisons of geometric mean titre ratios will be made utilizing the lower bound of two-sided score-based confidence intervals ($\alpha = 0.05$) with non-inferiority margin 0.67.

All non-inferiority comparisons of seroresponse rates will be made utilizing the lower bound of two-sided score-based confidence intervals ($\alpha = 0.05$) with non-inferiority margin -10%.

4.2.4 Geometric Mean Titres and Geometric Mean Fold Rise

Geometric mean titres (GMTs) and geometric mean fold rises (GMFRs) for antibody titres will be calculated for each treatment received and will be summarised at each scheduled visit as per protocol section 1.3. GMT and GMFR summaries will be based on each of the immunogenicity analysis sets defined in Section 3.2 of the master-SAP.

GMTs and GMFRs will be calculated using model-adjusted titre levels derived using an analysis of covariance (ANCOVA) model as described in Section 4.2.7. In addition, GMTs and GMFRs will be calculated using unadjusted titre levels.

Descriptive statistics for GMTs and GMFRs will include number of participants, geometric mean, 95% CI, minimum and maximum.

The GMT will be calculated as the antilogarithm of Σ(log2 transformed titre/n), i.e. as the antilogarithm transformation of the mean of the log-transformed titre, where n is the number of participants with titre information. The 95% CI about the GMT will be

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

calculated as the anti-logarithm transformation of the upper and lower limits for a two-sided CI for the mean of the log-transformed titres.

The fold rise is calculated as the ratio of the post-dose titre level to the pre-dose titre level. GMFR will be calculated as anti-logarithm of Σ (log2 transformed (post-dose titre/ pre-dose titre)/n). The 95% CIs for GMFR will be calculated similarly to those for GMT.

The statistical methodology will be based on a 2-sided 95% CI of the ratio of the GMTs. Non-inferiority will be demonstrated if the lower limit of the 2-sided 95% CI of the GMT ratio of the comparator group ($Group_C$) and the reference group ($Group_R$) and is >0.67. The 2-sided 95% CI for the ratio of GMTs will be calculated using normal approximation of log-transformed concentrations.

The 95% CI for the GMT ratio between 2 groups will be constructed as follows:

Logarithm transformation of the individual concentrations will be calculated.

The 95% CI for the difference in log(GMT) between 2 groups: $Group_C$ and $Group_R$ will be in the form:

$$\bar{X}_C - \bar{X}_R \pm t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right) \times S\sqrt{{}^{1}/_{n_C} + {}^{1}/_{n_R}}$$

Where $\bar{X}_C$ and $\bar{X}_R = \log(GMT)$ are the means of the log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$S^2 = {}^{[(n_C - 1)S_C^2 + (n_R - 1)S_R^2]}/_{(n_C + n_R - 2)}$ is the pooled sample variance,

n_C and n_R are the sample sizes for $Group_C$ and $Group_R$, respectively,

S_C and S_R are the sample standard deviations of log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right)$ is the $100\left(1 - \frac{\alpha}{2}\right)$ percentile of the t-distribution with degrees of freedom $df = n_C + n_R - 2$.

To test this hypothesis, a 2- sided 95% CI will be constructed around the ratio $\frac{GMT_C}{GMT_R}$, where GMT_C and GMT_R are the geometric mean of the antibody titres in the comparator and reference groups respectively, at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is > 0.67. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: {}^{GMT_C}/_{GMT_R} \leq 0.67$$

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

$$H_A: {}^{GMT_C}/_{GMT_R} > 0.67$$

Or equivalently

$$H_0: \log(GMT_C) - \log(GMT_R) \leq \log(0.67)$$

$$H_A: \log(GMT_C) - \log(GMT_R) > \log(0.67)$$

For the separately considered GMT hypotheses, if the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

4.2.5 Seroresponse Rate

Seroresponse is a binary outcome where a success is when the fold rise in titres compared to baseline is ≥4. Seroresponse will be calculated for each treatment group and will be summarised at each scheduled post-vaccination visit as for all titre measurements. Only participants with non-missing data at both baseline and the applicable post-baseline visit will be included in seroresponse calculations.

The number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method, will be provided.

The statistical methodology will be based on a 2-sided 95% CI of the difference in seroresponse rates. Non-inferiority will be demonstrated if the upper bound of the 2-sided 95% CI rate difference in seroresponse between the comparator group and reference group is ≥-10%. The 95% CI of the difference in proportions $P_C - P_R$ will be computed using the Newcombe score without continuity correction.

To test this hypothesis, a 2- sided 95% CI will be constructed around the difference $P_C - P_R$, where P_C and P_R are the proportions of participants in the comparator and reference groups respectively who are classified as seroresponders (≥4 fold increase from baseline) at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is ≥ -10%. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: P_C - P_R < -10\%$$

$$H_A: P_C - P_R \geq -10\%$$

If the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

4.2.6 Handling of Missing Data

The analysis of antibody geometric mean titres and geometric mean fold rise will use the following imputation method: a titre value measured below the lower limit of quantification (LLOQ) will be imputed to a value that is half of the LLOQ in summaries and analyses but will be listed as reported in the raw data. Titre values measured as above the upper limit of quantification (ULOQ) will be imputed at the ULOQ value.

4.2.7 Primary Analysis of Primary Endpoints

The primary endpoint is that 28 days after the second vaccination dose the GMT ratio of pseudoneutralizing antibodies against the B.1.351 variant elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval.

The estimand is expressed as the GMT ratio (GMT_{GroupC} / GMT_{GroupR}).

Primary analyses of GMT ratio will be performed on model-adjusted titre levels, which will be derived using an analysis of covariance (ANCOVA) model which includes the log transformed value of the titre as the dependent variable, and will include independent variables for the vaccine group, baseline co-morbidities, sex, and age group as fixed effects (see Section 4.2.7.1). Two-sided 95% CIs for GMT ratio will be obtained by anti-log the confidence limits for the adjusted mean difference of the logarithmically transformed assay results which are calculated as defined in Section 4.2.4.

4.2.7.1 Model Adjustment

Analyses of GMT/GMR will be performed on both unadjusted titre levels CCI CCI, as well as on model-adjusted titre levels CCI. The model-adjusted analyses will be derived using analysis of covariance (ANCOVA) models which are fitted on the pairwise comparisons given in Table 4, and include the log transformed value of the titre CCI as the dependent variable, and independent variables for vaccine group, visit window (baseline, Day 15, Day 29, Day 43, Day 57), baseline co-morbidities (Yes or No), sex (Male or Female) and age group (18-64 or 65 and older) as fixed effects. For pairwise comparisons within the same treatment arms, random terms for intercept and slope will be used to account for intrasubject correlation by strain. For pairwise comparisons between treatment arms, the analysis model will be similar to the model above, however, excluding grouping by strain. The least square means for the visit effect and their 95% CIs will be converted by anti-log into the adjusted GMT/GMR and its 95% CI at each visit.

For consistency with the modeling approach for the previously vaccinated cohorts, , model fitting for descriptive analyses will be performed by treatment group (i.e., not pairwise) utilizing only a random participant intercept.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Model adjustments will be performed on the seronegative analysis set only.

Seroresponse rates will also be analysed using the same method described above but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7. The model-adjusted titre levels will be derived as the LS means for each treatment group and visit combination, plus the residual from the model fit. The LS means will be obtained for a population with balanced groups of Male and Female, comorbidity status and age groups.

4.2.8 Descriptive Analysis of Primary Endpoints

Descriptive statistics for GMTs will include number of participants, geometric mean, 95% CI, minimum and maximum for each compared vaccine group.

4.2.9 Sensitivity Analyses of Primary Endpoint

Sensitivity analyses may explore the following:

- model adjustment using age as a continuous covariate (adjusting to the mean age across all participants)
- model adjustment including BMI and age as a continuous covariate (adjusting to the mean BMI and age across all participants)

Further sensitivity analyses may perform non-inferiority calculations on model adjusted values calculated from models fitting comparator arms simultaneously in order to provide a consistent effect of covariates across treatment arms.

4.2.10 Primary Analysis of Secondary Endpoints

4.2.10.1 GMT

The secondary endpoints (GMT) are detailed in Table 1 GMT ratio will be analyzed using a similar ANCOVA as the primary endpoints described in Section 4.2.7.

4.2.10.2 Seroresponse rate

The secondary endpoints (seroresponse rate) are detailed in Table 1.

Difference in seroresponse rate will be calculated and 2-sided 95% CI of the difference between vaccine groups will be computed using the Newcombe score without continuity correction, as described in Section 4.2.5.

Differences in seroresponse rates will also be analysed using the same method described above but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7 to calculate seroresponse.

4.2.11 Descriptive Analysis of Secondary Endpoint

Descriptive analyses for GMTs will be performed similarly to secondary analyses of the primary endpoint as described in Section 4.2.8.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Descriptive statistics for seroresponse rate will include the number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method.

4.2.12 Sensitivity Analyses of Secondary Endpoint

N/A

4.2.13 Exploratory Analyses of Primary and Secondary Endpoints

CCI

4.2.14 Comparisons

The primary and secondary immunogenicity objectives and the GMT and seroresponse comparisons for the previously unvaccinated participants receiving a 2-dose primary vaccination are presented in Table 4.

4.2.15 Planned Analyses

Table 2 Description of the Analysis Keys for Tables 3 – 4

Population	Analysis Key	Example
Previously unvaccinated	Vaccination treatment received: V1222 (2 doses of AZD1222) or V2816 (2 doses of AZD2816) or V1222/2816 (1 dose of AZD1222 followed by 1 dose of AZD2816) Dosing interval: (4): 4-week dosing interval or (12): 12-week dosing interval Strain: Wuhan (Wuhan-Hu-1) or Beta (Variant B.1.351) Analysis Timepoint: D1 (28 days post-dose 1) D2 (28 days post-dose 2)	[V1222 (4):Wuhan:D2] = Immunogenicity following primary vaccination with 2 doses of AZD1222 using a 4-week dosing interval against Wuhan-Hu-1 28 days post-dose 2

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Table 3 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)

Treatment	Interval	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	1	[V1222(4):Wuhan:D1]†
				Seroresponse	2	
			28 days after 2nd dose	GMT	3	[V1222(4):Wuhan:D2]
				Seroresponse	4	
		B.1.351	28 days after 1st dose	GMT	5	[V1222(4):Beta:D1]†
				Seroresponse	6	
			28 days after 2nd dose	GMT	7	[V1222(4):Beta:D2]
				Seroresponse	8	
AZD2816	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	9	[V2816(4):Wuhan:D1] ‡
				Seroresponse	10	
			28 days after 2nd dose	GMT	11	[V2816(4):Wuhan:D2]
				Seroresponse	12	
		B.1.351	28 days after 1st dose	GMT	13	[V2816(4):Beta:D1]‡
				Seroresponse	14	
			28 days after 2nd dose	GMT	15	[V2816(4):Beta:D2]
				Seroresponse	16	
AZD1222/2816	4 weeks	Wuhan-Hu-1	28 days after 2nd dose	GMT	17	[V1222/2816(4):Wuhan:D2]
				Seroresponse	18	
		B.1.351	28 days after 2nd dose	GMT	19	[V1222/2816(4):Beta:D2]
				Seroresponse	20	
AZD2816	12 weeks	Wuhan-Hu-1	28 days after 2nd dose	GMT	21	[V2816(12):Wuhan:D2]
				Seroresponse	22	
		B.1.351	28 days after 2nd dose	GMT	23	[V2816(12):Beta:D2]
				Seroresponse	24	

† descriptive summaries for 28 days after 1st dose will pool all treatment groups who received AZD1222 as their first dose (ie, homologous and heterologous series).
‡ descriptive summaries for 28 days after 1st dose will pool all treatment groups who received AZD2816 as their first dose (4-week interval and 12-week interval treatment arms).

GMT: Geometric mean titre

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Table 4 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups

Objective	$\frac{[\![\mathbf{[GMT]}]\!]_{\mathbf{comparator}}}{[\![\mathbf{[GMT]}]\!]_{\mathbf{reference}}}$	$[\![\mathbf{[Seroresponse]}]\!]_{\mathbf{comparator}} - [\![\mathbf{[Seroresponse]}]\!]_{\mathbf{reference}}$
To determine if the neutralizing antibody GMT response/seroresponse elicited by a 2-dose AZD2816 primary vaccination is non-inferior to the response elicited by a 2-dose AZD1222 primary vaccination	[V2816(4): Beta: D2]/[V1222(4): Wuhan: D2] (Co − Primary)	[V2816(4): Beta: D2] − [V1222(4): Wuhan: D2] (Co-primary)
	[V2816(4): Beta: D1]/[V1222(4): Wuhan: D1]	[V2816(4): Beta: D1] − [V1222(4): Wuhan: D1]
	$\frac{\text{[V2816(4): Beta: D2]}}{\text{[V1222(4): Beta: D2]}}$ (Key Secondary 2.1)	[V2816(4): Beta: D2] − [V1222(4): Beta: D2] (Other Secondary)
	[V2816(4): Beta: D1]/[V1222(4): Beta: D1]	[V2816(4): Beta: D1] − [V1222(4): Beta: D1]
	[V2816(4): Wuhan: D2]/[V1222(4): Wuhan: D2] (Key Secondary 2.3)	[V2816(4): Wuhan: D2] − [V1222(4): Wuhan: D2] (Other Secondary)
	[V2816(4): Wuhan: D1]/[V1222(4): Wuhan: D1]	[V2816(4): Wuhan: D1] − [V1222(4): Wuhan: D1]
To determine if the neutralizing antibody GMT response/seroresponse elicited by a 2-dose AZD1222 + AZD2816 heterologous primary vaccination is non-inferior to the response elicited by a 2-dose AZD1222 primary vaccination	[V1222/2816(4): Beta: D2] /[V1222(4): Wuhan: D2] (Key Secondary 2.2)	[V1222/2816(4): Beta: D2] − [V1222(4): Wuhan: D2] (Other Secondary)
	[V1222/2816(4): Wuhan: D2] /[V1222(4): Wuhan: D2](Other Secondary)	[V1222/2816(4): Wuhan: D2] − [V1222(4): Wuhan: D2](Other Secondary)
	[V1222/2816(4): Beta: D2] /[V1222(4): Beta: D2] (Other Secondary)	[V1222/2816(4): Beta: D2] − [V1222(4): Beta: D2] (Other Secondary)

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Table 4 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups

Objective	$\frac{[\![\mathbf{[GMT]}]\!]_{\mathbf{comparator}}]}{[\![\mathbf{[GMT]}]\!]_{\mathbf{reference}}]}$	$[\![\mathbf{[Seroresponse]}]\!]_{\mathbf{comparator}}] - [\![\mathbf{[Seroresponse]}]\!]_{\mathbf{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following a 2-dose AZD2816 primary vaccination	[V2816(4): Beta: D2]/[V2816(4): Wuhan: D2] (Other Secondary)	[V2816(4): Beta: D2] − [V2816(4): Wuhan: D2](Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following a 2-dose AZD1222/AZD2816 primary heterologous vaccination	[V1222/2816(4): Beta: D2]/[V1222 /2816(4): Wuhan: D2] (Other Secondary)	[V1222/2816(4): Beta: D2] − [V1222/2816(4): Wuhan: D2](Other Secondary)

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

4.2.16 Subgroup Analyses

Subgroup analyses will be performed on primary and secondary analyses within subgroups defined in Section 3.4 of the master-SAP.

4.2.17 Multiple comparisons

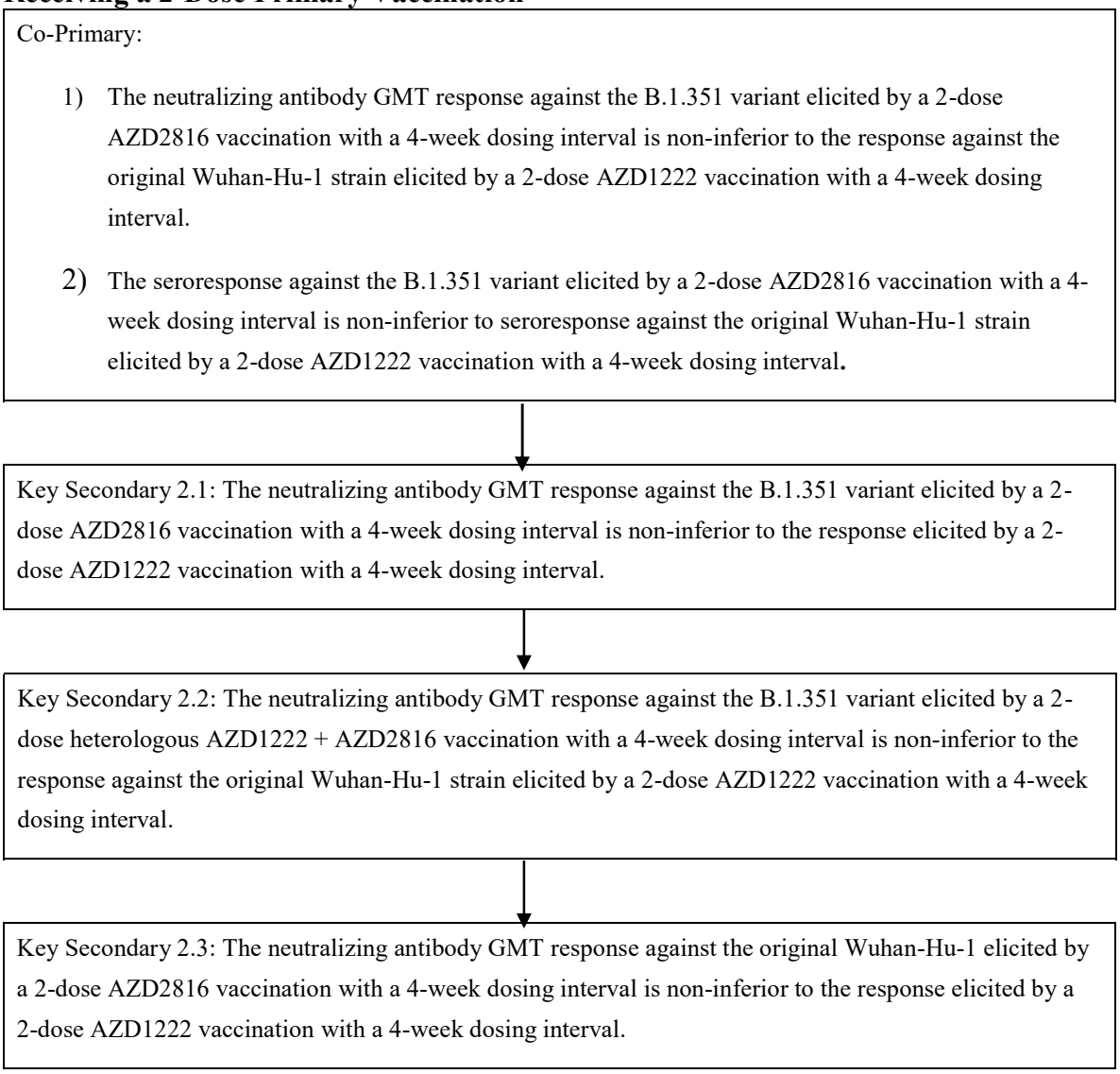
A hierarchical approach will be used to control for multiplicity of the primary and key secondary immunogenicity endpoints. That is, the null hypotheses for the immunogenicity endpoints will be tested in a hierarchical order, and the subsequent null hypothesis will be tested only if the prior null hypothesis is rejected. Study success for the primary endpoint will only be declared if non-inferiority criteria are met for both the GMT ratio and seroresponse criteria. Consequently, no adjustment to alpha for multiplicity will be made in the analysis of immune response. A separate hierarchies will be used for the previously unvaccinated cohort within this sub-SAP, with separate type I error rate controls. The primary statistical comparisons of safety data will not be adjusted for multiple comparisons.

The primary key endpoints will be analysed with the type I error rate of 5%. The key secondary endpoints will be analysed with the type I error rate of 5%, following the success of the primary objective. Following the success of the co primary objectives this study will conclude that the humoral immune response against the B.1.351 variant elicited by a 2-dose AZD2816 vaccination with a 4-week dosing interval is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by a 2-dose AZD1222 vaccination with a 4-week dosing interval. All other analyses for secondary endpoints and/or any further exploratory analyses for the primary and key secondary endpoint will be tested for nominal statistical significance only.

The testing procedure will continue down the hierarchy if the preceding endpoint is rejected at a two-sided 0.05 level and will stop if the preceding endpoint is not rejected at a two-sided 0.05 level. Statistical significance, using model-adjusted estimates, will be assessed in the following sequence:

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

Figure 1 Hypothesis Testing Order for Unvaccinated Seronegative Participants Receiving a 2-Dose Primary Vaccination



DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

4.3 Safety Analyses

Please refer to the master-SAP.

DocuSign Envelope ID: ADDE79DC-48B9-4EBB-A8F3-1274FCE4A1C9

5 REFERENCES

Little, R. J. A. and Rubin, D. B. Statistical Analysis with Missing Data, 2nd Edition, Hoboken, NJ: John Wiley & Sons 2002; 257.

Miettinen O, Nurminen M. Comparative analysis of two rates. Stat Med. 1985;4(2):213-26.

CHMP (Committee for Human Medicinal Products). Reflection paper on the regulatory requirements for vaccines intended to provide protection against variant strain(s) of SARS-CoV-2. https://www.ema.europa.eu/en/documents/scientific-guideline/reflection-paper-regulatory-requirements-vaccines-intended-provide-protection-against-variant_en.pdf. Published 2021.

Hing, J.P., Woolfrey, S.G., Greenslade, D. et al. Analysis of Toxicokinetic Data Using NONMEM: Impact of Quantification Limit and Replacement Strategies for Censored Data. J Pharmacokinet Pharmacodyn 28, 465–479 (2001). https://doi.org/10.1023/A:1012247131190

PPD

DocuSigned by PPD
PPD
I am the author of this document
23 September 2022 | 18:45 GMT
8C9818B7DD7449E0A7174A550A2608ED

DocuSign

Certificate Of Completion

Envelope Id: ADDE79DC48B94EBBA8F31274FCE4A1C9
Status: Completed
Subject: Please DocuSign: d7220c00001-naive-sap-ed-2.0.docx
Source Envelope:
Document Pages: 26
Signatures: 1
Envelope Originator:
Certificate Pages: 3
Initials: 0
PPD
AutoNav: Enabled
EnvelopeId Stamping: Enabled
Time Zone: (UTC+05:30) Chennai, Kolkata, Mumbai, New Delhi

Record Tracking

Status: Original
9/23/2022 11:17:00 PM
Holder: PPD
Location: DocuSign

Signer Events	Signature	Timestamp
PPD PPD Security Level: Email, Account Authentication (Required), Logged in	PPD Signature Adoption: Pre-selected Style Signature ID: 8C9818B7-DD74-49E0-A717-4A550A2608ED Using IP Address: 70.63.230.6 With Signing Authentication via DocuSign password With Signing Reasons (on each tab): I am the author of this document	Sent: 9/23/2022 11:25:34 PM Viewed: 9/24/2022 12:13:12 AM Signed: 9/24/2022 12:15:57 AM
Electronic Record and Signature Disclosure: Accepted: 9/24/2022 12:13:12 AM ID: a8641c22-d0ce-4ed9-88a5-20f806f055d0		

In Person Signer Events	Signature	Timestamp

Editor Delivery Events	Status	Timestamp

Agent Delivery Events	Status	Timestamp

Intermediary Delivery Events	Status	Timestamp

Certified Delivery Events	Status	Timestamp

Carbon Copy Events	Status	Timestamp

Witness Events	Signature	Timestamp

Notary Events	Signature	Timestamp

Envelope Summary Events	Status	Timestamps
Envelope Sent	Hashed/Encrypted	9/23/2022 11:25:34 PM
Certified Delivered	Security Checked	9/24/2022 12:13:12 AM
Signing Complete	Security Checked	9/24/2022 12:15:57 AM
Completed	Security Checked	9/24/2022 12:15:57 AM

Payment Events	Status	Timestamps

Electronic Record and Signature Disclosure

Electronic Record and Signature Disclosure created on: 1/31/2020 10:11:39 AM
Parties agreed to: PPD

I hereby consent to that AstraZeneca Worldwide https://www.astrazeneca.com may disclose personal information such as; full name, email address, and any other information you may supply on the electronic form to AstraZeneca affiliates and third party service providers throughout the world in relation to the handling and administration of the Electronic Signature Service solution. This consent relates to any electronic records or signatures associated with the electronic contract.
AstraZeneca and the third party administering this service store and process personal information that AstraZeneca collects from you for the purposes of operating the Electronic Signature Service solution.
This also applies after termination of the Agreement. Processing of your personal information will be done in accordance with applicable law.
You may request access to your personal data and withdraw agreement to this processing at any time by contacting us in writing at docusign-support@astrazeneca.com.
Personal details and electronic signatures of signatories contained in contracts cannot be removed once the contract has been executed and will remain part of such contracts until these are destroyed in accordance with applicable law and AstraZeneca internal data retention policies.

SIGNATURE PAGE

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature

Document Name: d7220c00001-naive-sap-ed-2		
Document Title:	D7220C00001 Naive Statistical Analysis Plan Edition 2	
Document ID:	Doc ID-004787736	
Version Label:	2.0 CURRENT LATEST APPROVED	
Server Date **(dd-MMM-yyyy HH:mm 'UTC'Z)**	**Signed by**	**Meaning of Signature**
23-Sep-2022 20:49 UTC	PPD	Author Approval
23-Sep-2022 23:24 UTC	PPD	Content Approval

Notes: (1) Document details as stored in ANGEL, an AstraZeneca document management system.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

STATISTICAL ANALYSIS PLAN	
Study Code	D7220C00001
Edition Number	2.0
Date	23-Sep-2022

A Phase II/III Partially Double-Blinded, Randomised, Multinational, Active-Controlled Study in Both Previously Vaccinated and Unvaccinated Adults to Determine the Safety and Immunogenicity of AZD2816, a Vaccine for the Prevention of COVID-19 Caused by Variant Strains of SARS-CoV-2

(Previous AZD1222 Cohort Sub-SAP)

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

TABLE OF CONTENTS

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

LIST OF TABLES

LIST OF FIGURES

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

LIST OF ABBREVIATIONS

Abbreviation or Specialized Term	Definition
AE	Adverse event
AESI	Adverse event of special interest
ANCOVA	Analysis of covariance
BMI	Body mass index
CI	Confidence Interval
CMI	Cell-mediated immune
COVID-19	Coronavirus 2019
CRF	Case report form
CSP	Clinical study protocol
GMFR	Geometric mean fold rises
GMT	Geometric mean titre
IMP	Investigational medicinal product
LLOQ	Lower limit of quantification
MAAE	Medically attended adverse event
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus 2
SD	Standard deviation
ULOQ	Upper limit of quantification

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

AMENDMENT HISTORY

CATEGORY* Change refers to:	Date	Description of change	In line with CSP?	Rationale
N/A		Initial approved SAP	N/A	N/A
Other	23 September, 2022	Deleted irrelevant reference, and added 2 new references	No	More complete documentation of methods

*Pre-specified categories are:

Primary or secondary endpoints; Statistical analysis method for the primary or secondary endpoints; Derivation of primary or secondary endpoints; Multiple Testing Procedure; Data presentations; Other

N/A = Not applicable

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

1 INTRODUCTION

The purpose of this document is to give details for the statistical analysis of study D7220C00001 supporting the clinical study report for participants previously vaccinated with AZD1222. The reader is referred to the Clinical Study Protocol (CSP), amendment 3 dated 11 October 2021 as well as the most recent versions of the Protocol Deviation Management Plan and the Case Report Form (CRF) for details of study conduct and data collection, and the master Statistical Analysis Plan (master-SAP) where referenced for details of the analysis principles appliable to all sub Statistical Analysis Plans (SAPs).

The term IMP (investigational medicinal product) is used throughout this SAP to include both treatment groups (AZD2816 and AZD1222).

1.1 Objectives and Endpoints

The primary safety objective for the cohort of seronegative previously vaccinated participants receiving a booster dose is to characterize the safety and tolerability of 1 booster dose of AZD2816 in participants previously vaccinated with AZD1222.

The primary and key secondary immunogenicity objectives for this cohort are as follows:

Primary:

- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

Key secondary:

- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.
- To determine if the neutralizing antibody GMT response elicited against the B.1.351 variant by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.
- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.
- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

The primary and key secondary immunogenicity objectives will be supported by other secondary objectives for which there will be no formal hypothesis testing.

Table 1 further describes the objectives and endpoints for this cohort of participants, including estimands for the primary and secondary immunogenicity objectives.

Table 1 Study Objectives and Endpoints for Participants Receiving a 1-Dose Booster Previously Vaccinated with AZD1222

Safety Objectives

Objectives	**Endpoints**
- Primary	
To characterize the safety and tolerability of 1 booster dose of AZD2816 in seronegative participants previously vaccinated with AZD1222	• Incidence of local and systemic solicited AEs for 7 days post dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs, for 28 days post-dose • The change from baseline for safety laboratory measures for 28 days post-dose
- Secondary	
To characterize the safety and tolerability of 1 booster dose of AZD1222 in seronegative participants previously vaccinated with AZD1222	• Incidence of local and systemic solicited AEs for 7 days post-dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs for 28 days post-dose
To characterize the extended safety of 1 booster dose of AZD2816 in seronegative participants previously vaccinated with AZD1222	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
To characterize the extended safety of 1 booster dose of AZD1222 in seronegative participants previously vaccinated with AZD1222	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
Immunogenicity objectives	
To determine if the neutralizing antibody GMT response elicited by an AZD1222 booster dose in patients previously vaccinated with AZD1222 is non-inferior to the response elicited by a 2-dose AZD1222 vaccination Estimand:	

Treatment	AZD1222 booster	AZD1222 primary vaccination
Population	AZD1222 vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective Level:	Serotype comparison:	

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Primary	Wuhan-Hu-1	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD1222 booster/AZD1222 vaccination	
To determine if the neutralizing antibody GMT response elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD2816 booster	AZD1222 primary vaccination
Population	AZD1222 vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Key Secondary 2.1	B.1.351	Wuhan-Hu-1
Key Secondary 2.5[a]	B.1.351	B.1.351
Key Secondary 2.3	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD2816 booster/AZD1222 vaccination	
To determine if the neutralizing antibody GMT response elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 Estimand:		
Treatment	AZD2816 booster	AZD1222 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Key Secondary 2.2	B.1.351	B.1.351
Key Secondary 2.4	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD2816 booster/AZD1222 booster	
To determine if the seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD2816 booster	AZD1222 primary vaccination
Population	AZD1222 vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 primary vaccination	
To determine if the seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 Estimand:		
Treatment	AZD2816 booster	AZD1222 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	B.1.351

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 booster	
To determine if the seroresponse elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD1222 booster	AZD1222 primary vaccination
Population	AZD1222 vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 primary vaccination	
To determine if the neutralizing antibody GMT response rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with AZD1222. Estimand:		
Treatment	AZD2816 booster	AZD2816 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Geometric mean titre ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the neutralizing antibody GMT response against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with AZD1222. Estimand:		
Treatment	AZD1222 booster	AZD1222 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Geometric mean titre ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with AZD1222. Estimand:		
Treatment	AZD2816 booster	AZD2816 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for B.1.351/Wuhan-Hu-1	

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with AZD1222. Estimand:		
Treatment	AZD1222 booster	AZD1222 booster
Population	AZD1222 vaccinated seronegative participants	AZD1222 vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for B.1.351/Wuhan-Hu-1	
To also determine non-inferiority of the s-protein binding antibody response for the above comparisons as other secondary objectives.		
To explore anti-vector responses to the ChAdOx-1 adenovirus vector following a booster dose of AZD2816 in sub-groups of seronegative and seropositive participants	• Magnitude of ChAdOx1 nAb titres (geometric mean titre) • Seroresponse rate of ChAdOx1 neutralizing antibody titres • Pairwise correlations between anti-S, pseudo-neutralization, and ChAdOx1 neutralizing antibody titres, 28 days after both Dose 1 and Dose 2	
Exploratory objectives		

CCI

[redacted]

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

CCI

[redacted]

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.

MAAEs: medically attended adverse events; SAEs: serious adverse events; AESIs: adverse events of special interest.

Seroresponse: An at least 4-fold increase in geometric mean titre from baseline.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

1.2 Study Design

Please refer to the master-SAP.

1.3 Sample Size

Please refer to the master-SAP.

2 CHANGES TO PROTOCOL PLANNED ANALYSES

By request of the Committee for Medicinal Products for Human Use (CHMP), separate SAPs have been created to analyze participants previously vaccinated with AZD1222, participants previously vaccinated with a mRNA vaccination, and previously unvaccinated participants. As a result, this SAP only specifies endpoints relevant to participants previously vaccinated with AZD1222.

For further details to changes to the protocol planned analysis please refer to the master-SAP.

3 DATA ANALYSIS CONSIDERATIONS

Please refer to the master-SAP.

4 STATISTICAL ANALYSIS

4.1 Study Population

Please refer to the master-SAP. This covers the cohort of participants previously vaccinated with AZD1222.

4.2 Immunogenicity Analyses

4.2.1 Humoral immune response following vaccination

For previously vaccinated participants, testing for antibody responses will be performed at baseline, day 15, day 29, and day 180.

For participants with a primary vaccination of AZD1222 with no booster, antibody response is collected at baseline, day 15, day 29, day 43 and day 57. Only participants with pseudoneutralising antibody titre assessments for both baseline and day 57.

The immunogenicity endpoints of interest in this study are:

- Geometric mean antibody titre
- Seroresponse, defined as ≥ 4-fold increase in the geometric mean antibody titre from baseline

Both the geometric mean antibody titre and seroresponse of participants will be summarised descriptively by strain, treatment received, and timepoint for the

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

immunogenicity analysis set. Scatter plots with box plot overlay for geometric mean antibody titre will be presented by strain, treatment received, and timepoint for the immunogenicity analysis set. Analyses will be performed for each of the following analysis sets:

- Immunogenicity analysis set
- Seronegative immunogenicity analysis set
- Seropositive immunogenicity analysis set

The following intercurrent events could impact the antibody levels achieved:

- receiving of immune-modifying drugs or vaccines
- SARS-CoV-2 infection during the study (COVID-19, SARS-CoV-2 test positive, asymptomatic COVID-19, COVID-19 pneumonia)

All immunogenicity descriptions and comparisons will use the principal stratum strategy, ie, all analyses will exclude data from participants that occur after any of the above intercurrent events.

The following antibody titre measurements from serum samples will be analysed:

- Spike-specific IgG response to SARS-CoV-2 by multiplexed immunoassay (S antibody)
- Antibody neutralisation using a lentivirus-based pseudovirus particle expressing the SARS-CoV-2 spike protein [neutralizing antibody (pseudoneutralization)]
- ChAdOx1 neutralizing antibody titres

CCI

Antibody responses will be tested against the following strains:

- Wuhan-Hu-1
- B.1.351 (beta)
- B.1.617.2 (delta)

Other variants of concern or interest may be analysed as exploratory analyses.

All assay results will be presented in units in which the assay is performed. Should an international standard be released, then immunogenicity analyses may be repeated using the international standards.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

4.2.2

CCI

CCI

4.2.3 Statistical Hypothesis

The overall hypothesis for participants previously vaccinated with 2-dose AZD1222 is that 28 days after a single booster dose, boosting with AZD1222 following a primary 2-dose vaccination series with AZD1222 will be non-inferior to a primary 2-dose vaccination series with AZD1222 in terms of immunogenicity.

All non-inferiority comparisons of geometric mean titre ratios will be made utilizing the lower bound of two-sided score-based 95% confidence intervals with non-inferiority margin 0.67.

All non-inferiority comparisons of seroresponse rates will be made utilizing the lower bound of two-sided score-based 95% confidence intervals with non-inferiority margin -10%.

4.2.4 Geometric Mean Titres and Geometric Mean Fold Rise

Geometric mean titres (GMTs) and geometric mean fold rises (GMFRs) for antibody titres will be calculated for each treatment received and will be summarised at each scheduled visit as per protocol section 1.3. GMT and GMFR summaries will be based on each of the immunogenicity analysis sets defined in Section 3.2 of the master-SAP.

GMTs and GMFRs will be calculated using model-adjusted titre levels derived using an analysis of covariance (ANCOVA) model as described in Section 4.2.7. In addition, GMTs and GMFRs will be calculated using unadjusted titre levels.

Descriptive statistics for GMTs and GMFRs will include number of participants, geometric mean, 95% CI, minimum and maximum.

The GMT will be calculated as the antilogarithm of Σ(log2 transformed titre/n), i.e. as the antilogarithm transformation of the mean of the log-transformed titre, where n is the number of participants with titre information. The 95% CI about the GMT will be calculated as the anti-logarithm transformation of the upper and lower limits for a two-sided CI for the mean of the log-transformed titres.

The fold rise is calculated as the ratio of the post-dose titre level to the pre-dose titre level. GMFR will be calculated as anti-logarithm of Σ (log2 transformed (post-dose titre/ pre-dose titre)/n). The 95% CIs for GMFR will be calculated similarly to those for GMT.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

The statistical methodology will be based on a 2-sided 95% CI of the ratio of the GMTs. Non-inferiority will be demonstrated if the lower limit of the 2-sided 95% CI of the GMT ratio of the comparator group ($Group_C$) and the reference group ($Group_R$) and is >0.67. The 2-sided 95% CI for the ratio of GMTs will be calculated using normal approximation of log-transformed concentrations.

The 95% CI for the GMT ratio between 2 groups will be constructed as follows:

Logarithm transformation of the individual concentrations will be calculated.

The 95% CI for the difference in log(GMT) between 2 groups: $Group_C$ and $Group_R$ will be in the form:

$$\bar{X}_C - \bar{X}_R \pm t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right) \times S\sqrt{{}^{1}/_{n_C} + {}^{1}/_{n_R}}$$

Where $\bar{X}_C$ and $\bar{X}_R = \log(GMT)$ are the means of the log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$S^2 = {}^{[(n_C - 1)S_C^2 + (n_R - 1)S_R^2]}/_{(n_C + n_R - 2)}$ is the pooled sample variance,

n_C and n_R are the sample sizes for $Group_C$ and $Group_R$, respectively,

S_C and S_R are the sample standard deviations of log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right)$ is the $100\left(1 - \frac{\alpha}{2}\right)$ percentile of the t-distribution with degrees of freedom $df = n_C + n_R - 2$.

To test this hypothesis, a 2- sided 95% CI will be constructed around the ratio $\frac{GMT_C}{GMT_R}$, where GMT_C and GMT_R are the geometric mean of the antibody titres in the comparator and reference groups respectively, at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is > 0.67. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: {}^{GMT_C}/_{GMT_R} \leq 0.67$$

$$H_A: {}^{GMT_C}/_{GMT_R} > 0.67$$

Or equivalently

$$H_0: \log(GMT_C) - \log(GMT_R) \leq \log(0.67)$$

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

$$H_A: \log(GMT_C) - \log(GMT_R) > \log(0.67)$$

For the separately considered GMT hypotheses, if the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

4.2.5 Seroresponse Rate

Seroresponse is a binary outcome where a success is when the fold rise in titres compared to baseline is ≥4. Seroresponse will be calculated for each treatment group and will be summarised at each scheduled post-vaccination visit as for all titre measurements. Only participants with non-missing data at both baseline and the applicable post-baseline visit will be included in seroresponse calculations.

The number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method, will be provided.

The statistical methodology will be based on a 2-sided 95% CI of the difference in seroresponse rates. Non-inferiority will be demonstrated if the upper bound of the 2-sided 95% CI rate difference in seroresponse between the comparator group and reference group is ≥-10%. The 95% CI of the difference in proportions $P_C - P_R$ will be computed using the Newcombe score without continuity correction.

To test this hypothesis, a 2- sided 95% CI will be constructed around the difference $P_C - P_R$, where P_C and P_R are the proportions of participants in the comparator and reference groups respectively who are classified as seroresponders (≥4 fold increase from baseline) at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is ≥ -10%. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: P_C - P_R < -10\%$$

$$H_A: P_C - P_R \geq -10\%$$

If the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

4.2.6 Handling of Missing Data

The analysis of antibody geometric mean titres and geometric mean fold rise will use the following imputation method: a titre value measured below the lower limit of quantification (LLOQ) will be imputed to a value that is half of the LLOQ in summaries and analyses but will be listed as reported in the raw data. Titre values measured as above the upper limit of quantification (ULOQ) will be imputed at the ULOQ value.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

4.2.7 Primary Analysis of Primary Endpoints

The primary endpoint is that the GMT ratio of pseudoneutralizing antibodies against the original Wuhan-Hu-1 strain elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222 28 days after booster is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants 28 days after second vaccination.

The estimand is expressed as the GMT ratio (GMT_{GroupC} / GMT_{GroupR}).

Primary analyses of GMT ratio will be performed on model-adjusted titre levels, which will be derived using an analysis of covariance (ANCOVA) model which includes the log transformed value of the titre as the dependent variable, and will include independent variables for the time since previous vaccination (for previously vaccinated individuals), baseline co-morbidities, sex, and age group as fixed effects (see Section 4.2.7.1) . Two-sided 95% CIs for GMT ratio will be obtained by anti-log the confidence limits for the adjusted mean difference of the logarithmically transformed assay results which are calculated as defined in Section 4.2.4.

4.2.7.1 Model Adjustment

Analyses of GMT/GMR will be performed on both unadjusted titre levels CCI CCI as well as on model-adjusted titre levels CCI CCI The model-adjusted analyses will be derived using analysis of covariance (ANCOVA) models which include the log transformed value of the titre CCI as the dependent variable, and independent variables for visit window (baseline, Day 15, Day 29, and additionally Day 43 and Day 57 for previously unvaccinated participants), baseline co-morbidities (Yes or No), sex (Male or Female) and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect. Additionally, for model adjustment for previously vaccinated cohorts time since previous vaccination will be included as a continuous (log-transformed) covariate. The least square means for the visit effect and their 95% CIs will be converted by anti-log into the adjusted GMT/GMR and its 95% CI at each visit.

For descriptive analyses, model fitting will be performed within each treatment group. Model adjustments will be performed on the seronegative analysis set only.

Model adjusted values will use the mean time since previous vaccination across previously vaccinated treatment arms (restricted to seronegative participants).

Seroresponse rates will also be analysed using the same method described above but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7. The model-adjusted titre levels will be derived as the LS means for each treatment group and visit combination, plus the residual from the model fit. The LS means will be obtained for a population with time since last vaccination, where appliable, set to

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

the mean observed such time, balanced groups of Male and Female, comorbidity status and age groups.

4.2.8 Descriptive Analysis of Primary Endpoints

Descriptive statistics for GMTs will include number of participants, geometric mean, 95% CI, minimum and maximum for each compared vaccine group.

4.2.9 Sensitivity Analyses of Primary Endpoint

Sensitivity analyses may explore the following:

- model adjustment using age as a continuous covariate (adjusting to the mean age across all participants in the primary analysis)
- model adjustment including BMI and age as a continuous covariate (adjusting to the mean BMI and age across all participants in the primary analysis).
- model adjustment including the dosing interval between the primary series vaccination for the previously vaccinated (adjusting for the mean dosing interval across all previously vaccinated participants in the primary analysis)
- a repeat of the primary and key secondary non-inferiority analyses utilising the in study AZD1222 primary series (4 week interval) as the reference arm instead of historic controls

4.2.10 Primary Analysis of Secondary Endpoints

4.2.10.1 GMT

The secondary endpoints (GMT) are detailed in Table 1. GMT ratio will be analyzed using a similar ANCOVA as the primary endpoints described in Section 4.2.7.

4.2.10.2 Seroresponse rate

The secondary endpoints (seroresponse rate) are detailed in Table 1.

Difference in seroresponse rate will be calculated and 2-sided 95% CI of the difference between vaccine groups will be computed using the Newcombe score without continuity correction, as described in Section 4.2.5.

Differences in seroresponse rates will also be analysed using the same method described above, but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7 to calculate seroresponse.

4.2.11 Descriptive Analysis of Secondary Endpoint

Descriptive analyses for GMTs will be performed similarly to secondary analyses of the primary endpoint as described in Section 4.2.8.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Descriptive statistics for seroresponse rate will include the number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method.

4.2.12 Sensitivity Analyses of Secondary Endpoint

N/A

4.2.13 Exploratory Analyses of Primary and Secondary Endpoints

CCI

[redacted]

4.2.14 Comparisons

The immunogenicity objectives and the GMT and seroresponse comparisons for the previously vaccinated participants receiving a 1-dose booster vaccination of AZD1222 are presented in Table 5.

Owing to national vaccine rollout in the recruitment countries, including the prioritization of elderly populations, it is anticipated that there will be critical differences between the previously vaccinated and previously unvaccinated cohorts with respect to age and presence of important underlying comorbidities that may confound the interpretation of the results.

Consequently, the primary and key secondary non-inferiority analyses across these two cohorts will compare the previously vaccinated participants that received a booster dose in this study with a subset of matched participants from the previously unvaccinated participants that received the 2-dose AZD1222 primary vaccine series in the AZD1222 Phase 3 study D8110C00001, which was performed in the US, Chile, and Peru.

This historical control group will be matched, at a minimum, to the previously vaccinated AZD1222 booster cohort in the D7220C00001 study based on age, BMI, gender, and presence of baseline comorbidities. These matched samples will then serve as the control arm for all planned non-inferiority analyses (both geometric mean titre [GMT] ratio and difference in seroresponse) of the previously vaccinated cohort treatment arms to the primary series vaccination.

Comparisons of antibody titres between the previously vaccinated cohort in this study and the historical controls from Study D8110C00001 will be conducted using the seronegative immunogenicity analysis described in Section 3.2 of the master-SAP, on the subset of historical control participants who had both pseudoneutralising titre assessments at both baseline and day 57 (ie, using an adjusted ANCOVA model to calculated adjusted means and standard errors for the historical comparators).

CCI

[redacted]

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

CCI

[redacted]

4.2.14.1.1 Historic Control Matching

Use one-to-one propensity score matching to match seronegative historical control participants from D8110c00001 in the immunogenicity analysis set (as defined in that study) who were previously vaccinated by a 2-dose AZD1222 vaccination (hence forth referred to as controls) to participants from this current study in the seronegative immunogenicity analysis set who received a booster dose of AZD2816 or AZD1222 and were previously vaccinated with AZD1222, prior to unbinding (hence forth referred to as cases).

Lists of control subjects to be provided based on adjustment of

i. Sex
ii. Age
iii. Baseline co-morbidities
iv. Baseline BMI.

Selection of controls using covariates (i)-(iv) is based upon blinded D7220c00001 data, including possible interactions, by assessing the relationship to study membership (either D8110c00001 or D7220c00001), with no assessment on the relationship of these covariates to the response. For example, this could be done by forward or backwards selection when fitting a logistic regression on study membership.

Once the covariates have been selected a one-to-one full matching algorithm will be used to provide the list of matched controls from D8110c00001.

Exact matching will be done with respect to factor variables.

A caliper will be considered to ensure a common support region is established for the covariates from each study. Plots will be used to investigate the bias-variance trade-off for different caliper sizes, with the aim of retaining the one-to-one ratio whilst improving balance between covariates. Note that as a matched sample generally has lower variability then bias reduction should be favored.

Alternate distance metrics will be explored during the matching (e.g. the propensity score, the logit of the propensity score, or Mahalanobis distance derived from one of these two adjusting for continuous covariates).

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Note that the use of weights is not applicable with a one-to-one matching as the same number of cases and controls are used in each matched sample.

The list of subjects identified as the list of controls from the D8110c00001 study will be stored as a note to file prior to database lock.

Based on this list of controls the subset of participants who received both primary vaccinations of AZD1222, had pseudo neutralizing antibody assessments at baseline and day 57, and day 57 had no SARS-CoV-2 infection during the study or were not the receipt of any immune-modifying drugs, blood products, or vaccines, will be selected. This will ensure that the population of historical controls are selected consistently with the seronegative immunogenicity analysis set defined in Section 3.2 of the master-SAP, along with the same principal stratum strategy applied to account for intercurrent events as described in Section 4.2.1, but with the additional constraint of having pseudo neutralizing antibody assessments at baseline and day 57 (thus ensuring a consistent number of subjects presented for the comparisons of GMT ratio and difference in seroresponse).

4.2.15 Planned Analyses

Table 2 Description of the Analysis Keys for Tables 3 – 5

Population	Analysis Key	Example
Previously unvaccinated	Vaccination treatment received: V1222 (2 doses of AZD1222) or V2816 (2 doses of AZD2816) or HV1222: ([historical] 2 doses of AZD1222 from study D8110C00001) Dosing interval: (4): 4-week dosing interval Strain: Wuhan (Wuhan-Hu-1) or Beta (Variant B.1.351) Analysis Timepoint: D1 (28 days post-dose 1) D2 (28 days post-dose 2)	[V1222 (4):Wuhan:D2] = Immunogenicity following primary vaccination with 2 doses of AZD1222 using a 4-week dosing interval against Wuhan-Hu-1 28 days post-dose 2
Previously vaccinated	Pre-study primary vaccination: V1222 (2 doses of AZD1222) Treatment received: B1222 (1 booster dose of AZD1222) or B2816 (1 booster dose of AZD2816) Strain: Wuhan (Wuhan-Hu-1) or Beta (Variant B.1.351) Note: analysis timepoint is 28 days post-booster dose	[V1222:B1222:Beta] = Immunogenicity in participants who were previously vaccinated with 2 doses of AZD1222 as primary vaccination series and received a single boost dose of AZD1222 against the B.1.351 variant

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 3 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)- Reference Groups

Treatment	Interval	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	4 weeks	B.1.351	28 days after 1st dose	GMT	1	[V1222(4):Beta:D1]
				Seroresponse	2	
			28 days after 2nd dose	GMT	3	[V1222(4):Beta:D2]
				Seroresponse	4	
AZD1222 (Historical Controls)	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	5	[HV1222(4):Wuhan:D1]
				Seroresponse	10	
			28 days after 2nd dose	GMT	11	[HV1222(4):Wuhan:D2]
				Seroresponse	12	

GMT: Geometric mean titre

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 4 Immunogenicity Descriptive Analysis for Previously Vaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)

Primary vaccination	Booster Treatment	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	AZD1222	Wuhan-Hu-1	28 days after booster dose	GMT	1	[V1222:B1222:Wuhan]
				Seroresponse	2	
		B.1.351	28 days after booster dose	GMT	3	[V1222:B1222:Beta]
				Seroresponse	4	
	AZD2816	Wuhan-Hu-1	28 days after booster dose	GMT	5	[V1222:B2816:Wuhan]
				Seroresponse	6	
		B.1.351	28 days after booster dose	GMT	7	[V1222:B2816:Beta]
				Seroresponse	8	

GMT: Geometric mean titre

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 5 Immunogenicity Comparisons for Previously AZD1222 Vaccinated Group

Objective	$\frac{[\![\text{GMT}]\!]_{\text{comparator}}]}{[\![\text{GMT}]\!]_{\text{reference}}]}$	$[\![\text{Seroresponse}]\!]_{\text{comparator}}] - [\![\text{Seroresponse}]\!]_{\text{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD1222 booster dose in patients previously vaccinated with AZD1222 is non-inferior to the response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{V1222: B1222: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Primary)	$[\text{V1222: B1222: Wuhan}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
	$\frac{[\text{V1222: B1222: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Other Secondary)[a]	$[\text{V1222: B1222: Beta}] - [\text{V1222(4): Beta: D2}]$ (Other Secondary)[a]
	$\frac{[\text{V1222: B1222: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Other Secondary)	$[\text{V1222: B1222: Beta}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{V1222: B2816: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.1)	$[\text{V1222: B2816: Beta}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
	$\frac{[\text{V1222: B2816: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Key Secondary 2.5)[a]	$[\text{V1222: B2816: Beta}] - [\text{V1222(4): Beta: D2}]$ (Other Secondary)[a]
	$\frac{[\text{V1222: B2816: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.3)	$[\text{V1222: B2816: Wuhan}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222	$\frac{[\text{V1222: B2816: Beta}]}{[\text{V1222: B1222: Beta}]}$ (Key Secondary 2.2)	$[\text{V1222: B2816: Beta}] - [\text{V1222: B1222: Beta}]$ (Other Secondary)
	$\frac{[\text{V1222: B2816: Wuhan}]}{[\text{V1222: B1222: Wuhan}]}$ (Key Secondary 2.4)	$[\text{V1222: B2816: Wuhan}] - [\text{V1222: B1222: Wuhan}]$ (Other Secondary)

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with AZD1222	$\frac{[V1222: B2816: Beta]}{[V1222: B2816: Wuhan]}$ (Other Secondary)	$[V1222: B2816: Beta] - [V1222: B2816: Wuhan]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with AZD1222	$\frac{[V1222: B1222: Beta]}{[V1222: B1222: Wuhan]}$ (Other Secondary)	$[V1222: B1222: Beta] - [V1222: B1222: Wuhan]$ (Other Secondary)

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

4.2.16 Subgroup Analyses

Subgroup analyses will be performed on primary and secondary analyses within subgroups defined in Section 3.4 of the master-SAP.

4.2.17 Multiple comparisons

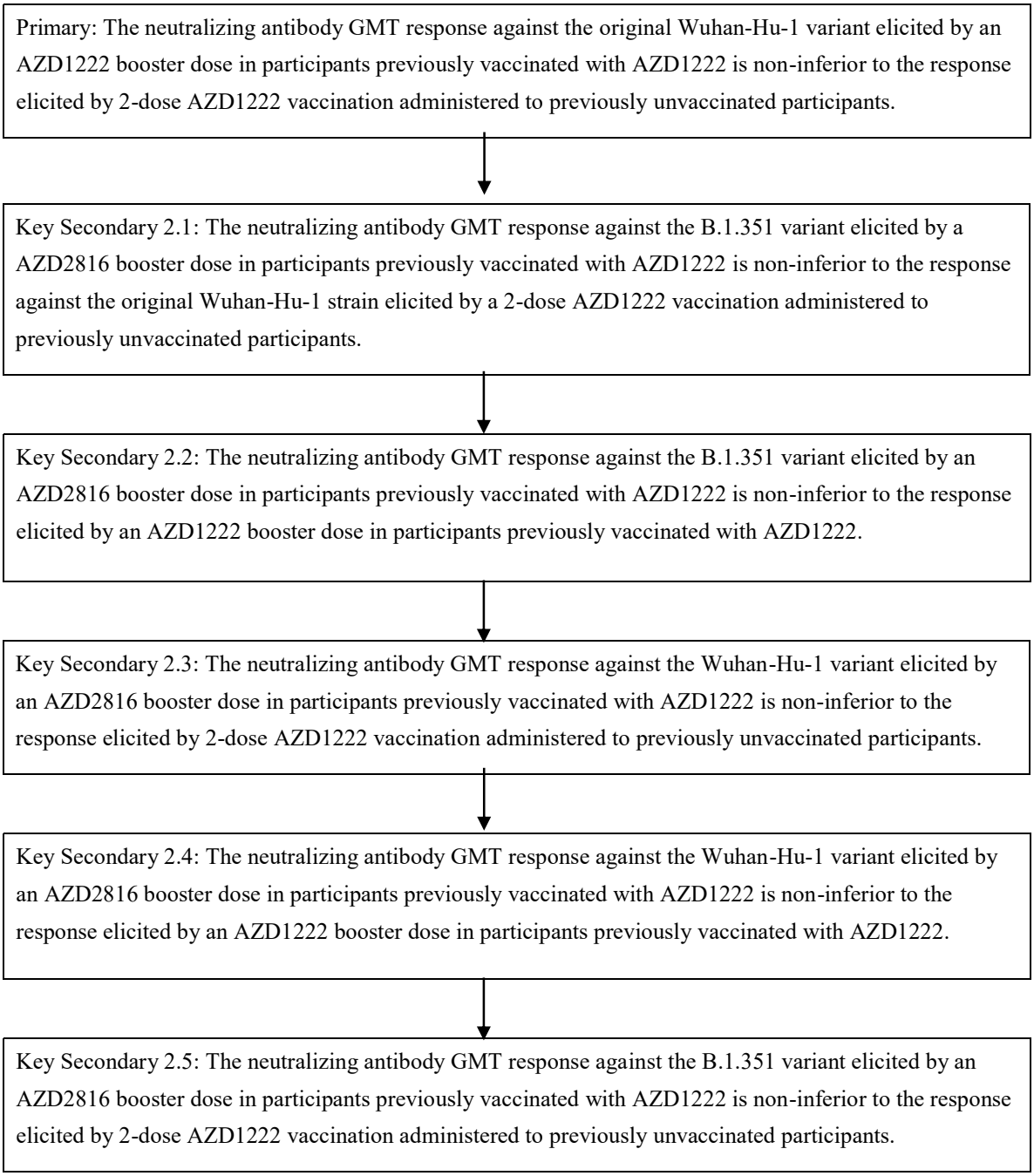
A hierarchical approach will be used to control for multiplicity of the primary and key secondary immunogenicity endpoints. That is, the null hypotheses for the immunogenicity endpoints will be tested in a hierarchical order, and the subsequent null hypothesis will be tested only if the prior null hypothesis is rejected. Consequently, no adjustment to alpha for multiplicity will be made in the analysis of immune response. Separate hierarchies will be used for the previously vaccinated cohort receiving a 2-dose primary vaccination series with AZD1222 within this sub-SAP, with separate type I error rate controls. The primary statistical comparisons of safety data will not be adjusted for multiple comparisons.

The primary key endpoints will be analysed with the type I error rate of 5%. The key secondary endpoints will be analysed with the type I error rate of 5%, following the success of the primary objective. All other analyses for secondary endpoints and/or any further exploratory analyses for the primary and key secondary endpoint will be tested for nominal statistical significance only.

The testing procedure will continue down the hierarchy if the preceding endpoint is rejected at a two-sided 0.05 level and will stop if the preceding endpoint is not rejected at a two-sided 0.05 level. Statistical significance, using model-adjusted estimates, will be assessed in the following sequence:

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Figure 1 Hypothesis Testing Order for Previously Vaccinated Seronegative Participants Receiving a 1-Dose Booster



DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

4.3 Safety Analyses

Please refer to master-SAP.

5 INTERIM ANALYSIS

Details of the initial interim analyses are provided in the Interim Analysis Charter.

The second interim analysis is to be performed for the data cut-off on the 11 October 2021 for the analyses not greyed-out from **Table 6**, **Table 7** and **Table 8**.

Table 6 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)- Reference Groups at Interim

Treatment	Interval	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	4 weeks	B.1.351	28 days after 1st dose	GMT	1	[V1222(4):Beta:D1]
				Seroresponse	2	
			28 days after 2nd dose	GMT	3	[V1222(4):Beta:D2]
				Seroresponse	4	
AZD1222 (Historical Controls)	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	5	[HV1222(4):Wuhan:D1]
				Seroresponse	10	
			28 days after 2nd dose	GMT	11	[HV1222(4):Wuhan:D2]
				Seroresponse	12	

GMT: Geometric mean titre
Tables in grey are not planned to be performed at the interim but will be carried out during the primary analysis.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 7 Immunogenicity Descriptive Analysis for Previously Vaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses) – Interim

Primary vaccination	Booster Treatment	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	AZD1222	Wuhan-Hu-1	28 days after booster dose	GMT	1	[V1222:B1222:Wuhan]
				Seroresponse	2	
		B.1.351	28 days after booster dose	GMT	3	[V1222:B1222:Beta]
				Seroresponse	4	
	AZD2816	Wuhan-Hu-1	28 days after booster dose	GMT	5	[V1222:B2816:Wuhan]
				Seroresponse	6	
		B.1.351	28 days after booster dose	GMT	7	[V1222:B2816:Beta]
				Seroresponse	8	

GMT: Geometric mean titre

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 8 Immunogenicity Comparisons for Previously AZD1222 Vaccinated Group – Interim

Objective	$\frac{[\![\text{GMT}]\!]_{\text{comparator}}]}{[\![\text{GMT}]\!]_{\text{reference}}]}$	$[\![\text{Seroresponse}]\!]_{\text{comparator}}] - [\![\text{Seroresponse}]\!]_{\text{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD1222 booster dose in patients previously vaccinated with AZD1222 is non-inferior to the response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{V1222: B1222: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Primary)	[V1222: B1222: Wuhan] − [HV1222(4): Wuhan: D2] (Other Secondary)
	$\frac{[\text{V1222: B1222: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Other Secondary)[a]	[V1222: B1222: Beta] − [V1222(4): Beta: D2] (Other Secondary)[a]
	$\frac{[\text{V1222: B1222: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Other Secondary)	[V1222: B1222: Beta] − [HV1222(4): Wuhan: D2] (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with AZD1222 is non-inferior to response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{V1222: B2816: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.1)	[V1222: B2816: Beta] − [HV1222(4): Wuhan: D2] (Other Secondary)
	$\frac{[\text{V1222: B2816: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Key Secondary 2.5)[a]	[V1222: B2816: Beta] − [V1222(4): Beta: D2] (Other Secondary)[a]
	$\frac{[\text{V1222: B2816: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.3)	[V1222: B2816: Wuhan] − [HV1222(4): Wuhan: D2] (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222	$\frac{[\text{V1222: B2816: Beta}]}{[\text{V1222: B1222: Beta}]}$ (Key Secondary 2.2)	[V1222: B2816: Beta] − [V1222: B1222: Beta] (Other Secondary)
	$\frac{[\text{V1222: B2816: Wuhan}]}{[\text{V1222: B1222: Wuhan}]}$ (Key Secondary 2.4)	[V1222: B2816: Wuhan] − [V1222: B1222: Wuhan] (Other Secondary)

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

Table 8 Immunogenicity Comparisons for Previously AZD1222 Vaccinated Group – Interim

Objective	$\frac{[\![\mathbf{[GMT]}]\!]_{\mathbf{comparator}}]}{[\![\mathbf{[GMT]}]\!]_{\mathbf{reference}}]}$	$[\![\mathbf{[Seroresponse]}]\!]_{\mathbf{comparator}}] - [\![\mathbf{[Seroresponse]}]\!]_{\mathbf{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose	$\frac{[\text{V1222: B2816: Beta}]}{[\text{V1222: B2816: Wuhan}]}$ (Other Secondary)	$[\text{V1222: B2816: Beta}] - [\text{V1222: B2816: Wuhan}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose	$\frac{[\text{V1222: B1222: Beta}]}{[\text{V1222: B1222: Wuhan}]}$ (Other Secondary)	$[\text{V1222: B1222: Beta}] - [\text{V1222: B1222: Wuhan}]$ (Other Secondary)

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.
GMT: Geometric mean titre
Tables in grey are not planned to be performed at the interim but will be carried out during the primary analysis.

DocuSign Envelope ID: BFF70BB7-7963-4B2D-809D-927C64E31A7F

6 REFERENCES

Little, R. J. A. and Rubin, D. B. Statistical Analysis with Missing Data, 2nd Edition, Hoboken, NJ: John Wiley & Sons 2002; 257.

Miettinen O, Nurminen M. Comparative analysis of two rates. Stat Med. 1985;4(2):213-26.

CHMP (Committee for Human Medicinal Products). Reflection paper on the regulatory requirements for vaccines intended to provide protection against variant strain(s) of SARS-CoV-2. https://www.ema.europa.eu/en/documents/scientific-guideline/reflection-paper-regulatory-requirements-vaccines-intended-provide-protection-against-variant_en.pdf. Published 2021.

Hing, J.P., Woolfrey, S.G., Greenslade, D. et al. Analysis of Toxicokinetic Data Using NONMEM: Impact of Quantification Limit and Replacement Strategies for Censored Data. J Pharmacokinet Pharmacodyn 28, 465–479 (2001). https://doi.org/10.1023/A:1012247131190

Due to technical issues, PPD (PPD PPD) (ClinChoice) was not able to sign in ANGEL/CARA, but is signing instead in DocuSign.

DocuSigned by PPD
PPD
I am the author of this document
23 September 2022 | 18:41 GMT
8C9818B7DD7449E0A7174A550A2608ED

DocuSign

Certificate Of Completion

Envelope Id: BFF70BB779634B2D809D927C64E31A7F
Status: Completed
Subject: Please DocuSign: d7220c00001-v1222-sap-ed-2.0.docx
Source Envelope:
Document Pages: 33
Signatures: 1
Certificate Pages: 3
Initials: 0
AutoNav: Enabled
EnvelopeId Stamping: Enabled
Time Zone: (UTC+05:30) Chennai, Kolkata, Mumbai, New Delhi

Envelope Originator:
PPD
PPD
IP Address: 24.30.87.232

Record Tracking

Status: Original
9/23/2022 11:27:40 PM
Holder: PPD
PPD
Location: DocuSign

Signer Events	Signature	Timestamp
PPD PPD Security Level: Email, Account Authentication (Required), Logged in	PPD Signature Adoption: Pre-selected Style Signature ID: 8C9818B7-DD74-49E0-A717-4A550A2608ED Using IP Address: 70.63.230.6 With Signing Authentication via DocuSign password With Signing Reasons (on each tab): I am the author of this document	Sent: 9/23/2022 11:28:54 PM Viewed: 9/24/2022 12:10:58 AM Signed: 9/24/2022 12:11:23 AM
Electronic Record and Signature Disclosure: Accepted: 9/24/2022 12:10:58 AM ID: 72a39eca-2291-4d7f-a391-18097400da33		

In Person Signer Events	Signature	Timestamp

Editor Delivery Events	Status	Timestamp

Agent Delivery Events	Status	Timestamp

Intermediary Delivery Events	Status	Timestamp

Certified Delivery Events	Status	Timestamp

Carbon Copy Events	Status	Timestamp

Witness Events	Signature	Timestamp

Notary Events	Signature	Timestamp

Envelope Summary Events	Status	Timestamps
Envelope Sent	Hashed/Encrypted	9/23/2022 11:28:54 PM
Certified Delivered	Security Checked	9/24/2022 12:10:58 AM
Signing Complete	Security Checked	9/24/2022 12:11:23 AM
Completed	Security Checked	9/24/2022 12:11:23 AM

Payment Events	Status	Timestamps

Electronic Record and Signature Disclosure

Electronic Record and Signature Disclosure created on: 1/31/2020 10:11:39 AM
Parties agreed to: PPD

I hereby consent to that AstraZeneca Worldwide https://www.astrazeneca.com may disclose personal information such as; full name, email address, and any other information you may supply on the electronic form to AstraZeneca affiliates and third party service providers throughout the world in relation to the handling and administration of the Electronic Signature Service solution. This consent relates to any electronic records or signatures associated with the electronic contract.
AstraZeneca and the third party administering this service store and process personal information that AstraZeneca collects from you for the purposes of operating the Electronic Signature Service solution.
This also applies after termination of the Agreement. Processing of your personal information will be done in accordance with applicable law.
You may request access to your personal data and withdraw agreement to this processing at any time by contacting us in writing at docusign-support@astrazeneca.com.
Personal details and electronic signatures of signatories contained in contracts cannot be removed once the contract has been executed and will remain part of such contracts until these are destroyed in accordance with applicable law and AstraZeneca internal data retention policies.

SIGNATURE PAGE

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature

Document Name: d7220c00001-v1222-sap-ed-2		
Document Title:	D7220C00001 V1222 Statistical Analysis Plan Edition 2	
Document ID:	Doc ID-004787738	
Version Label:	2.0 CURRENT LATEST APPROVED	
Server Date **(dd-MMM-yyyy HH:mm 'UTC'Z)**	**Signed by**	**Meaning of Signature**
23-Sep-2022 20:46 UTC	PPD	Author Approval
23-Sep-2022 23:25 UTC	PPD	Content Approval

Notes: (1) Document details as stored in ANGEL, an AstraZeneca document management system.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

STATISTICAL ANALYSIS PLAN	
Study Code	D7220C00001
Edition Number	2.0
Date	23-Sep-2022

A Phase II/III Partially Double-Blinded, Randomised, Multinational, Active-Controlled Study in Both Previously Vaccinated and Unvaccinated Adults to Determine the Safety and Immunogenicity of AZD2816, a Vaccine for the Prevention of COVID-19 Caused by Variant Strains of SARS-CoV-2 – (Previous mRNA Cohort Sub-SAP)

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

TABLE OF CONTENTS

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

LIST OF TABLES

LIST OF FIGURES

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

LIST OF ABBREVIATIONS

Abbreviation or Specialized Term	Definition
AE	Adverse event
AESI	Adverse event of special interest
ANCOVA	Analysis of covariance
BMI	Body mass index
CI	Confidence Interval
CMI	Cell-mediated immune
COVID-19	Coronavirus 2019
CRF	Case report form
CSP	Clinical study protocol
GMFR	Geometric mean fold rises
GMT	Geometric mean titre
IMP	Investigational medicinal product
LLOQ	Lower limit of quantification
MAAE	Medically attended adverse event
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus 2
SD	Standard deviation
ULOQ	Upper limit of quantification

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

AMENDMENT HISTORY

CATEGORY* Change refers to:	Date	Description of change	In line with CSP?	Rationale
N/A	01 February, 2022	Initial approved SAP	N/A	N/A
Other	23 September, 2022	Deleted irrelevant reference, and added 2 new references	No	More complete documentation of methods

*Pre-specified categories are:

Primary or secondary endpoints; Statistical analysis method for the primary or secondary endpoints; Derivation of primary or secondary endpoints; Multiple Testing Procedure; Data presentations; Other

N/A = Not applicable

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

1 INTRODUCTION

The purpose of this document is to give details for the statistical analysis of study D7220C00001 supporting the clinical study report for the cohort of participants previously vaccinated with an mRNA primary series. The reader is referred to the Clinical Study Protocol (CSP), amendment 3 dated 11 October 2021 as well as the most recent versions of the Protocol Deviation Management Plan, and the Case Report Form (CRF) for details of study conduct and data collection, and the master Statistical Analysis Plan (master-SAP) where referenced for details of the analysis principles appliable to all sub Statistical Analysis Plans (SAPs).

The term IMP (investigational medicinal product) is used throughout this SAP to include both treatment groups (AZD2816 and AZD1222).

1.1 Objectives and Endpoints

The primary safety objective for the cohort of seronegative previously vaccinated participants receiving a booster dose is to characterize the safety and tolerability of 1 booster dose of AZD2816 in participants previously vaccinated with an mRNA primary series vaccination.

The primary and key secondary immunogenicity objectives for this cohort are as follows:

Primary:

- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

Key secondary:

- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine is non-inferior to the response against the original Wuhan-Hu-1 strain elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.
- To determine if the neutralizing antibody GMT response elicited against the B.1.351 variant by an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine.
- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD2816 booster dose in participants previously

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

vaccinated with a mRNA vaccine is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

- To determine if the neutralizing antibody GMT response against the original Wuhan-Hu-1 strain elicited by an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine.
- To determine if the neutralizing antibody GMT response against the B.1.351 variant elicited by an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants.

The primary and key secondary immunogenicity objectives will be supported by other secondary objectives for which there will be no formal hypothesis testing.

Table 1 further describes the objectives and endpoints for this cohort of participants, including estimands for the primary and secondary immunogenicity objectives.

Table 1 Study Objectives and Endpoints for Participants Receiving a 1-Dose Booster Previously Vaccinated with mRNA vaccine

Safety Objectives

Objectives	**Endpoints**
- Primary	
To characterize the safety and tolerability of 1 booster dose of AZD2816 in seronegative participants previously vaccinated with an mRNA vaccine	• Incidence of local and systemic solicited AEs for 7 days post dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs, for 28 days post-dose • The change from baseline for safety laboratory measures for 28 days post-dose
- Secondary	
To characterize the safety and tolerability of 1 booster dose of AZD1222 in seronegative participants previously vaccinated with an mRNA vaccine	• Incidence of local and systemic solicited AEs for 7 days post-dose • Incidence of unsolicited AEs, including MAAEs, SAEs, and AESIs for 28 days post-dose
To characterize the extended safety of 1 booster dose of AZD2816 in seronegative participants previously vaccinated with an mRNA vaccine	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
To characterize the extended safety of 1 booster dose of AZD1222 in seronegative participants previously vaccinated with an mRNA vaccine	• Incidence of MAAEs, SAEs, and AESIs from Day 1 through 6 months post-vaccination
Immunogenicity objectives	
To determine if the neutralizing antibody GMT response elicited by an AZD1222 booster dose in patients previously vaccinated with an mRNA vaccine is non-inferior to the response elicited by a 2-dose AZD1222 vaccination Estimand:	

Treatment	AZD1222 booster	AZD1222 primary vaccination

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Population	mRNA vaccine vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective Level:	Serotype comparison:	
Primary	Wuhan-Hu-1	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD1222 booster/AZD1222 vaccination	
To determine if the neutralizing antibody GMT response elicited by an AZD2816 booster dose in participants previously vaccinated with an mRNA vaccine is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD2816 booster	AZD1222 primary vaccination
Population	mRNA vaccine vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Key Secondary 2.1	B.1.351	Wuhan-Hu-1
Key Secondary 2.5[a]	B.1.351	B.1.351
Key Secondary 2.3	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD2816 booster/AZD1222 vaccination	
To determine if the neutralizing antibody GMT response elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with an mRNA vaccine Estimand:		
Treatment	AZD2816 booster	AZD1222 booster
Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Key Secondary 2.2	B.1.351	B.1.351
Key Secondary 2.4	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	GMT ratio of pseudoneutralizing antibodies for AZD2816 booster/AZD1222 booster	
To determine if the seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with an mRNA vaccine is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD2816 booster	AZD1222 primary vaccination
Population	mRNA vaccine vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 primary vaccination	
To determine if the seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with an mRNA vaccine Estimand:		
Treatment	AZD2816 booster	AZD1222 booster

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	B.1.351
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 booster	
To determine if the seroresponse elicited by an AZD1222 booster dose in participants previously vaccinated with an mRNA vaccine is non-inferior to response elicited by a 2-dose AZD1222 vaccination Estimand:		
Treatment	AZD1222 booster	AZD1222 primary vaccination
Population	mRNA vaccine vaccinated seronegative participants	Seronegative participants with no prior COVID-19 vaccination
Timepoint	28 days after booster administration	28 days after second vaccination dose
Objective level:	Serotype comparison:	
Other Secondary	Wuhan-Hu-1	Wuhan-Hu-1
Other Secondary[a]	B.1.351	B.1.351
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for AZD2816 booster versus AZD1222 primary vaccination	
To determine if the neutralizing antibody GMT response rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with an mRNA vaccine. Estimand:		
Treatment	AZD2816 booster	AZD2816 booster
Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Geometric mean titre ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the neutralizing antibody GMT response against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with an mRNA vaccine. Estimand:		
Treatment	AZD1222 booster	AZD1222 booster
Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Geometric mean titre ratio of pseudoneutralizing antibodies for B.1.351/Wuhan-Hu-1	
To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with an mRNA vaccine. Estimand:		
Treatment	AZD2816 booster	AZD2816 booster

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for B.1.351/Wuhan-Hu-1	
To determine if the seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with an mRNA vaccine. Estimand:		
Treatment	AZD1222 booster	AZD1222 booster
Population	mRNA vaccine vaccinated seronegative participants	mRNA vaccine vaccinated seronegative participants
Timepoint	28 days after booster administration	28 days after booster administration
Objective level:	Serotype comparison:	
Other Secondary	B.1.351	Wuhan-Hu-1
Endpoint	Difference in seroresponse (≥4-fold increase from baseline in pseudoneutralizing antibodies) for B.1.351/Wuhan-Hu-1	
To also determine non-inferiority of the s-protein binding antibody response for the above comparisons as other secondary objectives.		
To explore anti-vector responses to the ChAdOx-1 adenovirus vector following a booster dose of AZD2816 in sub-groups of seronegative and seropositive participants	• Magnitude of ChAdOx1 nAb titres (geometric mean titre) • Seroresponse rate of ChAdOx1 neutralizing antibody titres • Pairwise correlations between anti-S, pseudo-neutralization, and ChAdOx1 neutralizing antibody titres, 28 days after both Dose 1 and Dose 2	
Exploratory objectives		

CCI

[redacted]

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

CCI

[redacted]

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.
MAAEs: medically attended adverse events; SAEs: serious adverse events; AESIs: adverse events of special interest.
Seroresponse: An at least 4-fold increase in geometric mean titre from baseline.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

1.2 Study Design

Please refer to the master-SAP.

1.3 Sample Size

Please refer to the master-SAP.

2 CHANGES TO PROTOCOL PLANNED ANALYSES

By request of the European Medicines Agency (EMA), separate SAPs have been created to analyze participants previously vaccinated with AZD1222, participants previously vaccinated with a mRNA vaccination, and previously unvaccinated participants. As a result, this SAP only specifies endpoints relevant to participants previously vaccinated with a mRNA vaccine.

For further details to changes to the protocol planned analysis following feedback from the EMA and Medicines and Healthcare products Regulatory Agency (MHRA) please refer to the master-SAP.

3 DATA ANALYSIS CONSIDERATIONS

Please refer to the master-SAP.

4 STATISTICAL ANALYSIS

4.1 Study Population

Please refer to the master-SAP. This covers the cohort of participants previously vaccinated with a mRNA vaccine.

4.2 Immunogenicity Analyses

4.2.1 Humoral immune response following vaccination

For previously vaccinated participants, testing for antibody responses will be performed at baseline, day 15, day 29, and day 180.

For participants with a primary vaccination of AZD1222 with no booster, antibody response is collected at baseline, day 15, day 29, day 43 and day 57. Only participants with titre assessments for both baseline and day 57.

The immunogenicity endpoints of interest in this study are:

- Geometric mean antibody titre
- Seroresponse, defined as ≥ 4-fold increase in the geometric mean antibody titre from baseline

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Both the geometric mean antibody titre and seroresponse of participants will be summarised descriptively by strain, treatment received, and timepoint for the immunogenicity analysis set. Scatter plots with box plot overlay for geometric mean antibody titre will be presented by strain, treatment received, and timepoint for the immunogenicity analysis set. Analyses will be performed for each of the following analysis sets:

- Immunogenicity analysis set
- Seronegative immunogenicity analysis set
- Seropositive immunogenicity analysis set

The following intercurrent events could impact the antibody levels achieved:

- receiving of immune-modifying drugs or vaccines
- SARS-CoV-2 infection during the study (COVID-19, SARS-CoV-2 test positive, asymptomatic COVID-19, COVID-19 pneumonia)

All immunogenicity descriptions and comparisons will use the principal stratum strategy, ie, all analyses will exclude data from participants that occur after any of the above intercurrent events.

The following antibody titre measurements from serum samples will be analysed:

- Spike-specific IgG response to SARS-CoV-2 by multiplexed immunoassay (S antibody)
- Antibody neutralisation using a lentivirus-based pseudovirus particle expressing the SARS-CoV-2 spike protein [neutralizing antibody (pseudoneutralization)]
- ChAdOx1 neutralizing antibody titres

CCI

Antibody responses will be tested against the following strains:

- Wuhan-Hu-1
- B.1.351 (beta)
- B.1.617.2 (delta)

Other variants of concern or interest may be analysed as exploratory analyses.

All assay results will be presented in units in which the assay is performed. Should an international standard be released, then immunogenicity analyses may be repeated using the international standards.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

4.2.2 CCI

CCI

4.2.3 Statistical Hypothesis

The overall hypothesis for participants previously vaccinated with a 2-dose mRNA vaccine is that 28 days after a single booster dose, boosting with AZD1222 following a primary 2-dose vaccination series with a mRNA vaccine will be non-inferior to a primary 2-dose vaccination series with AZD1222 in terms of immunogenicity.

All non-inferiority comparisons of geometric mean titre ratios will be made utilizing the lower bound of two-sided score-based confidence intervals ($\alpha = 0.05$) with non-inferiority margin 0.67.

All non-inferiority comparisons of seroresponse rates will be made utilizing the lower bound of two-sided score-based confidence intervals ($\alpha = 0.05$) with non-inferiority margin -10%.

4.2.4 Geometric Mean Titres and Geometric Mean Fold Rise

Geometric mean titres (GMTs) and geometric mean fold rises (GMFRs) for antibody titres will be calculated for each treatment received and will be summarised at each scheduled visit as per protocol section 1.3. GMT and GMFR summaries will be based on each of the immunogenicity analysis sets defined in Section 3.2 of the master-SAP

GMTs and GMFRs will be calculated using model-adjusted titre levels derived using an analysis of covariance (ANCOVA) model as described in Section 4.2.7. In addition, GMTs and GMFRs will be calculated using unadjusted titre levels.

Descriptive statistics for GMTs and GMFRs will include number of participants, geometric mean, 95% CI, minimum and maximum.

The GMT will be calculated as the antilogarithm of Σ(log2 transformed titre/n), i.e. as the antilogarithm transformation of the mean of the log-transformed titre, where n is the number of participants with titre information. The 95% CI about the GMT will be calculated as the anti-logarithm transformation of the upper and lower limits for a two-sided CI for the mean of the log-transformed titres.

The fold rise is calculated as the ratio of the post-dose titre level to the pre-dose titre level. GMFR will be calculated as anti-logarithm of Σ (log2 transformed (post-dose titre/ pre-dose titre)/n). The 95% CIs for GMFR will be calculated similarly to those for GMT.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

The statistical methodology will be based on a 2-sided 95% CI of the ratio of the GMTs. Non-inferiority will be demonstrated if the lower limit of the 2-sided 95% CI of the GMT ratio of the comparator group ($Group_C$) and the reference group ($Group_R$) and is >0.67. The 2-sided 95% CI for the ratio of GMTs will be calculated using normal approximation of log-transformed concentrations.

The 95% CI for the GMT ratio between 2 groups will be constructed as follows:

Logarithm transformation of the individual concentrations will be calculated.

The 95% CI for the difference in log(GMT) between 2 groups: $Group_C$ and $Group_R$ will be in the form:

$$\bar{X}_C - \bar{X}_R \pm t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right) \times S\sqrt{{}^{1}/_{n_C} + {}^{1}/_{n_R}}$$

Where $\bar{X}_C$ and $\bar{X}_R = \log(GMT)$ are the means of the log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$S^2 = {}^{[(n_C - 1)S_C^2 + (n_R - 1)S_R^2]}/_{(n_C + n_R - 2)}$ is the pooled sample variance,

n_C and n_R are the sample sizes for $Group_C$ and $Group_R$, respectively,

S_C and S_R are the sample standard deviations of log-transformed concentration for $Group_C$ and $Group_R$, respectively,

$t\left(1 - {}^{\alpha}/_{2}, n_C + n_R - 2\right)$ is the $100\left(1 - \frac{\alpha}{2}\right)$ percentile of the t-distribution with degrees of freedom $df = n_C + n_R - 2$.

To test this hypothesis, a 2- sided 95% CI will be constructed around the ratio $\frac{GMT_C}{GMT_R}$, where GMT_C and GMT_R are the geometric mean of the antibody titres in the comparator and reference groups respectively, at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is > 0.67. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: {}^{GMT_C}/_{GMT_R} \leq 0.67$$

$$H_A: {}^{GMT_C}/_{GMT_R} > 0.67$$

Or equivalently

$$H_0: \log(GMT_C) - \log(GMT_R) \leq \log(0.67)$$

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

$$H_A: \log(GMT_C) - \log(GMT_R) > \log(0.67)$$

For the separately considered GMT hypotheses, if the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

4.2.5 Seroresponse Rate

Seroresponse is a binary outcome where a success is when the fold rise in titres compared to baseline is ≥4. Seroresponse will be calculated for each treatment group and will be summarised at each scheduled post-vaccination visit as for all titre measurements. Only participants with non-missing data at both baseline and the applicable post-baseline visit will be included in seroresponse calculations.

The number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method, will be provided.

The statistical methodology will be based on a 2-sided 95% CI of the difference in seroresponse rates. Non-inferiority will be demonstrated if the upper bound of the 2-sided 95% CI rate difference in seroresponse between the comparator group and reference group is ≥-10%. The 95% CI of the difference in proportions $P_C - P_R$ will be computed using the Newcombe score without continuity correction.

To test this hypothesis, a 2- sided 95% CI will be constructed around the difference $P_C - P_R$, where P_C and P_R are the proportions of participants in the comparator and reference groups respectively who are classified as seroresponders (≥4 fold increase from baseline) at the timepoints post vaccination for which the groups are being compared.

The hypothesis will be supported by the data, if the lower bound of the calculated 95% CI is ≥ -10%. This is equivalent to testing the null hypothesis using a 1-sided type-I error rate of 0.025.

$$H_0: P_C - P_R < -10\%$$

$$H_A: P_C - P_R \geq -10\%$$

If the null hypothesis is rejected, then the alternative hypothesis of non-inferiority will be supported.

4.2.6 Handling of Missing Data

The analysis of antibody geometric mean titres and geometric mean fold rise will use the following imputation method: a titre value measured below the lower limit of quantification (LLOQ) will be imputed to a value that is half of the LLOQ in summaries and analyses, but will be listed as reported in the raw data. Titre values measured as above the upper limit of quantification (ULOQ) will be imputed at the ULOQ value.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

4.2.7 Primary Analysis of Primary Endpoints

The primary endpoint is that the GMT ratio of pseudoneutralizing antibodies against the original Wuhan-Hu-1 strain elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine 28 days after booster is non-inferior to the response elicited by 2-dose AZD1222 vaccination administered to previously unvaccinated participants 28 days after second vaccination.

The estimand is expressed as the GMT ratio (GMT_{GroupC} / GMT_{GroupR}).

Primary analyses of GMT ratio will be performed on model-adjusted titre levels, which will be derived using an analysis of covariance (ANCOVA) model which includes the log transformed value of the titre as the dependent variable, and will include independent variables for the time since previous vaccination (for previously vaccinated individuals), baseline co-morbidities, sex, and age group as fixed effects (see Section 4.2.7.1). Two-sided 95% CIs for GMT ratio will be obtained by anti-log the confidence limits for the adjusted mean difference of the logarithmically transformed assay results which are calculated as defined in Section 4.2.4.

4.2.7.1 Model Adjustment

Analyses of GMT/GMR will be performed on both unadjusted titre levels CCI CCI as well as on model-adjusted titre levels CCI CCI. The model-adjusted analyses will be derived using analysis of covariance (ANCOVA) models which include the log transformed value of the titre or CCI as the dependent variable, and independent variables for visit window (baseline, Day 15, Day 29, and additionally Day 43 and Day 57 for previously unvaccinated participants), baseline co-morbidities (Yes or No), sex (Male or Female) and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect. Additionally, for model adjustment for previously vaccinated cohorts time since previous vaccination will be included as a continuous (log-transformed) covariate. The least square means for the visit effect and their 95% CIs will be converted by anti-log into the adjusted GMT/GMR and its 95% CI at each visit.

For descriptive analyses, model fitting will be performed within each treatment group. Model adjustments will be performed on the seronegative analysis set only.

Model adjusted values will use the mean time since previous vaccination across previously vaccinated treatment arms (restricted to seronegative participants).

Seroresponse rates will also be analysed using the same method described above but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7. The model-adjusted titre levels will be derived as the LS means for each treatment group and visit combination, plus the residual from the model fit. The LS means will be obtained for a population with time since last vaccination, where applicable, set to

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

the mean observed such time, balanced groups of Male and Female, comorbidity status and age groups.

4.2.8 Descriptive Analysis of Primary Endpoints

Descriptive statistics for GMTs will include number of participants, geometric mean, 95% CI, minimum and maximum for each compared vaccine group.

4.2.9 Sensitivity Analyses of Primary Endpoint

Sensitivity analyses may explore the following:

- model adjustment using age as a continuous covariate (adjusting to the mean age across all participants in the primary analysis)
- model adjustment including BMI and age as a continuous covariate (adjusting to the mean BMI and age across all participants in the primary analysis).
- model adjustment including the dosing interval between the primary series vaccination for the previously vaccinated (adjusting for the mean dosing interval across all previously vaccinated participants in the primary analysis)

4.2.10 Primary Analysis of Secondary Endpoints

4.2.10.1 GMT

The secondary endpoints (GMT) are detailed in Table 1. GMT ratio will be analyzed using a similar ANCOVA as the primary endpoints described in Section 4.2.7.

4.2.10.2 Seroresponse rate

The secondary endpoints (seroresponse rate) are detailed Table 1.

Difference in seroresponse rate will be calculated and 2-sided 95% CI of the difference between vaccine groups will be computed using the Newcombe score without continuity correction, as described in Section 4.2.5.

Differences in seroresponse rates will also be analysed using the same method described above, but using the model-adjusted baseline and post-baseline titre levels from the model described in Section 4.2.7 to calculate seroresponse.

4.2.11 Descriptive Analysis of Secondary Endpoint

Descriptive analyses for GMTs will be performed similarly to secondary analyses of the primary endpoint as described in Section 4.2.8.

Descriptive statistics for seroresponse rate will include the number and percentage of participants with post-vaccination seroresponse, and 95% CIs, calculated using the Clopper-Pearson exact method.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

4.2.12 Sensitivity Analyses of Secondary Endpoint

N/A

4.2.13 Exploratory Analyses of Primary and Secondary Endpoints

CCI

[redacted]

4.2.14 Comparisons

The immunogenicity objectives and the GMT and seroresponse comparisons for the previously vaccinated participants receiving a 1-dose booster vaccination are presented in **Table 5.**

Owing to national vaccine rollout in the recruitment countries, including the prioritization of elderly populations, it is anticipated that there will be critical differences between the previously vaccinated and previously unvaccinated cohorts with respect to age and presence of important underlying comorbidities that may confound the interpretation of the results.

Consequently, the primary and key secondary non-inferiority analyses across these two cohorts will compare the previously vaccinated participants that received a booster dose in this study with a subset of matched participants from the previously unvaccinated participants that received the 2-dose AZD1222 primary vaccine series in the AZD1222 Phase 3 study D8110C00001, which was performed in the US, Chile, and Peru.

This historical control group will be matched, at a minimum, to the previously vaccinated AZD1222 booster cohort in the D7220C00001 study based on age, BMI, gender, and presence of baseline comorbidities. These matched samples will then serve as the control arm for all planned non-inferiority analyses (both geometric mean titre [GMT] ratio and difference in seroresponse) of the previously vaccinated cohort treatment arms to the primary series vaccination.

Comparisons of antibody titres between the previously vaccinated cohort in this study and the historical controls from Study D8110C00001 will be conducted using the seronegative immunogenicity analysis described in Section 3.2 of the master-SAP, on the subset of historical control participants who had both pseudoneutralising titre assessments at both baseline and day 57 (ie, using an adjusted ANCOVA model to calculated adjusted means and standard errors for the historical comparators).

CCI

[redacted]

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

CCI

4.2.14.1.1 Historic Control Matching

Use one-to-one propensity score matching to match seronegative historical control participants from D8110c00001 in the immunogenicity analysis set who were previously vaccinated by a 2-dose AZD1222 vaccination (hence forth referred to as controls) to participants from this current study in the seronegative immunogenicity analysis set who received a booster dose of AZD2816 or AZD1222 and were previously vaccinated with AZD1222, prior to unbinding (hence forth referred to as cases).

Lists of control subjects to be provided based on adjustment of

i. Sex
ii. Age
iii. Baseline co-morbidities
iv. Baseline BMI.

Selection of controls using covariates (i)-(iv) is based upon blinded D7220c00001 data, including possible interactions, by assessing the relationship to study membership (either D8110c00001 or D7220c00001), with no assessment on the relationship of these covariates to the response. For example, this could be done by forward or backwards selection when fitting a logistic regression on study membership.

Once the covariates have been selected a one-to-one full matching algorithm will be used to provide the list of matched controls from D8110c00001.

Exact matching will be done with respect to factor variables.

A caliper will be considered to ensure a common support region is established for the covariates from each study. Plots will be used to investigate the bias-variance trade-off for different caliper sizes, with the aim of retaining the one-to-one ratio whilst improving balance between covariates. Note that as a matched sample generally has lower variability then bias reduction should be favored.

Alternate distance metrics will be explored during the matching (e.g. the propensity score, the logit of the propensity score, or Mahalanobis distance derived from one of these two adjusting for continuous covariates).

Note that the use of weights is not applicable with a one-to-one matching as the same number of cases and controls are used in each matched sample.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

The list of subjects identified as the list of controls from the D8110c00001 study will be stored as a note to file prior to database lock.

Based on this list of controls the subset of participants who received both primary vaccinations of AZD1222, had pseudo neutralizing antibody assessments at baseline and day 57, and day 57 had no SARS-CoV-2 infection during the study or were not the receipt of any immune-modifying drugs, blood products, or vaccines, will be selected. This will ensure that the population of historical controls are selected consistently with the seronegative immunogenicity analysis set defined in Section 3.2 of the master-SAP, along with the same principal stratum strategy applied to account for intercurrent events as described in Section 4.2.1, but with the additional constraint of having pseudo neutralizing antibody assessments at baseline and day 57 (thus ensuring a consistent number of subjects presented for the comparisons of GMT ratio and difference in seroresponse).

4.2.15 Planned Analyses

Table 2 Description of the Analysis Keys for Tables 3 – 6

Population	Analysis Key	Example
Previously unvaccinated	Vaccination treatment received: V1222 (2 doses of AZD1222) or V2816 (2 doses of AZD2816) or HV1222: ([historical] 2 doses of AZD1222 from study D8110C00001) Dosing interval: (4): 4-week dosing interval Strain: Wuhan (Wuhan-Hu-1) or Beta (Variant B.1.351) Analysis Timepoint: D1 (28 days post-dose 1) D2 (28 days post-dose 2)	[V1222 (4):Wuhan:D2] = Immunogenicity following primary vaccination with 2 doses of AZD1222 using a 4-week dosing interval against Wuhan-Hu-1 28 days post-dose 2
Previously vaccinated	Pre-study primary vaccination: VmRNA (2 doses of an mRNA vaccine) Treatment received: B1222 (1 booster dose of AZD1222) or B2816 (1 booster dose of AZD2816) Strain: Wuhan (Wuhan-Hu-1) or Beta (Variant B.1.351) Note: analysis timepoint is 28 days post-booster dose	[VmRNA:B1222:Beta] = Immunogenicity in participants who were previously vaccinated with 2 doses of a mRNA vaccine as primary vaccination series and received a single boost dose of AZD1222 against the B.1.351 variant

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 3 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)- Reference Groups

Treatment	Interval	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	4 weeks	B.1.351	28 days after 1st dose	GMT	1	[V1222(4):Beta:D1]
				Seroresponse	2	
			28 days after 2nd dose	GMT	3	[V1222(4):Beta:D2]
				Seroresponse	4	
AZD1222 (Historical Controls)	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	5	[HV1222(4):Wuhan:D1] ‡
				Seroresponse	6	
			28 days after 2nd dose	GMT	7	[HV1222(4):Wuhan:D2]
				Seroresponse	8	

GMT: Geometric mean titre

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 4 Immunogenicity Descriptive Analysis for Previously Vaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)

Primary vaccination	Booster Treatment	Strain	Timepoint	Endpoint	Index	Analysis Key
mRNA	AZD1222	Wuhan-Hu-1	28 days after booster dose	GMT	1	[VmRNA:B1222:Wuhan]
				Seroresponse	2	
		B.1.351	28 days after booster dose	GMT	3	[VmRNA:B1222:Beta]
				Seroresponse	4	
	AZD2816	Wuhan-Hu-1	28 days after booster dose	GMT	5	[VmRNA:B2816:Wuhan]
				Seroresponse	6	
		B.1.351	28 days after booster dose	GMT	7	[VmRNA:B2816:Beta]
				Seroresponse	8	

GMT: Geometric mean titre

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 5 Immunogenicity Comparisons for Previously mRNA Vaccinated Group

Objective	$\frac{[\![\text{GMT}]\!]_{\text{comparator}}]}{[\![\text{GMT}]\!]_{\text{reference}}]}$	$[\![\text{Seroresponse}]\!]_{\text{comparator}}] - [\![\text{Seroresponse}]\!]_{\text{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD1222 booster dose in patients previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{VmRNA: B1222: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Primary)	$[\text{VmRNA: B1222: Wuhan}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
	$\frac{[\text{VmRNA: B1222: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Other Secondary)[a]	$[\text{VmRNA: B1222: Beta}] - [\text{V1222(4): Beta: D2}]$ (Other Secondary)[a]
	$\frac{[\text{VmRNA: B1222: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Other Secondary)	$[\text{VmRNA: B1222: Beta}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with a mRNA is non-inferior to response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.1)	$[\text{VmRNA: B2816: Beta}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Key Secondary 2.5)[a]	$[\text{VmRNA: B2816: Beta}] - [\text{V1222(4): Beta: D2}]$ (Other Secondary)[a]
	$\frac{[\text{VmRNA: B2816: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.3)	$[\text{VmRNA: B2816: Wuhan}] - [\text{HV1222(4): Wuhan: D2}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{VmRNA: B1222: Beta}]}$ (Key Secondary 2.2)	$[\text{VmRNA: B2816: Beta}] - [\text{VmRNA: B1222: Beta}]$ (Other Secondary)
	$\frac{[\text{VmRNA: B2816: Wuhan}]}{[\text{VmRNA: B1222: Wuhan}]}$ (Key Secondary 2.4)	$[\text{VmRNA: B2816: Wuhan}] - [\text{VmRNA: B1222: Wuhan}]$ (Other Secondary)

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 5 Immunogenicity Comparisons for Previously mRNA Vaccinated Group

Objective	$\frac{[\![\mathbf{[GMT]}]\!]_{\mathbf{comparator}}]}{[\![\mathbf{[GMT]}]\!]_{\mathbf{reference}}]}$	$[\![\mathbf{[Seroresponse]}]\!]_{\mathbf{comparator}}] - [\![\mathbf{[Seroresponse]}]\!]_{\mathbf{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{VmRNA: B2816: Wuhan}]}$ (Other Secondary)	$[\text{VmRNA: B2816: Beta}] - [\text{VmRNA: B2816: Wuhan}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B1222: Beta}]}{[\text{VmRNA: B1222: Wuhan}]}$ (Other Secondary)	$[\text{VmRNA: B1222: Beta}] - [\text{VmRNA: B1222: Wuhan}]$ (Other Secondary)

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

4.2.16 Subgroup Analyses

Subgroup analyses will be performed on primary and secondary analyses within subgroups defined in Section 3.4 of the master-SAP.

4.2.17 Multiple comparisons

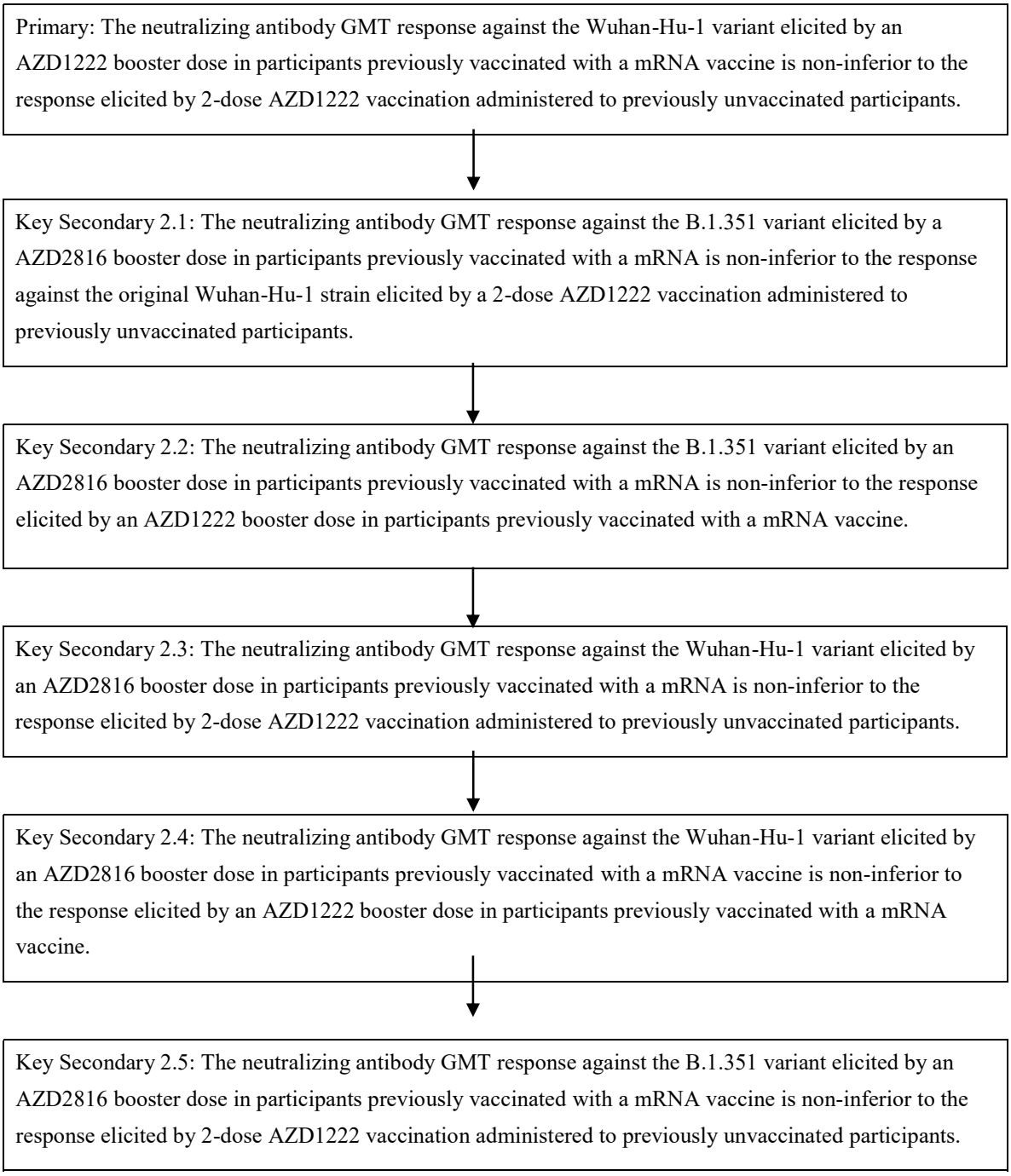
A hierarchical approach will be used to control for multiplicity of the primary and key secondary immunogenicity endpoints. That is, the null hypotheses for the immunogenicity endpoints will be tested in a hierarchical order, and the subsequent null hypothesis will be tested only if the prior null hypothesis is rejected. Consequently, no adjustment to alpha for multiplicity will be made in the analysis of immune response. Separate hierarchies will be used for the previously vaccinated cohort receiving a 2-dose primary vaccination series with a mRNA vaccine within this sub-SAP, with separate type I error rate controls. The primary statistical comparisons of safety data will not be adjusted for multiple comparisons.

The primary key endpoints will be analysed with the type I error rate of 5%. The key secondary endpoints will be analysed with the type I error rate of 5%, following the success of the primary objective. All other analyses for secondary endpoints and/or any further exploratory analyses for the primary and key secondary endpoint will be tested for nominal statistical significance only.

The testing procedure will continue down the hierarchy if the preceding endpoint is rejected at a two-sided 0.05 level and will stop if the preceding endpoint is not rejected at a two-sided 0.05 level. Statistical significance, using model-adjusted estimates, will be assessed in the following sequence:

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Figure 1 Hypothesis Testing Order for Previously Vaccinated Seronegative Participants Receiving a 1-Dose Booster



DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

4.3 Safety Analyses

Please refer to master-SAP.

5 INTERIM ANALYSIS

Details of the initial interim analysis are provided in the Interim Analysis Charter.

The second interim analysis is to be performed for the data cut-off on the 11 October 2021 for the analyses not greyed-out from **Table 6**, **Table 7** and **Table 8**.

Table 6 Immunogenicity Descriptive Analysis for Previously Unvaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses)- Reference Groups for Interim Analysis

Treatment	Interval	Strain	Timepoint	Endpoint	Index	Analysis Key
AZD1222	4 weeks	B.1.351	28 days after 1st dose	GMT	1	[V1222(4):Beta:D1]
				Seroresponse	2	
			28 days after 2nd dose	GMT	3	[V1222(4):Beta:D2]
				Seroresponse	4	
AZD1222 (Historical Controls)	4 weeks	Wuhan-Hu-1	28 days after 1st dose	GMT	5	[HV1222(4):Wuhan:D1] [t]
				Seroresponse	60	
			28 days after 2nd dose	GMT	7	[HV1222(4):Wuhan:D2]
				Seroresponse	8	

GMT: Geometric mean titre
Tables in grey are not planned to be performed at the interim but will be carried out during the primary analysis.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 7 Immunogenicity Descriptive Analysis for Previously Vaccinated Treatment Groups (with Separate Seronegative and Seropositive Analyses) – Interim Analysis

Primary vaccination	Booster Treatment	Strain	Timepoint	Endpoint	Index	Analysis Key
mRNA vaccine	AZD1222	Wuhan-Hu-1	28 days after booster dose	GMT	1	[VmRNA:B1222:Wuhan]
				Seroresponse	2	
		B.1.351	28 days after booster dose	GMT	3	[VmRNA:B1222:Beta]
				Seroresponse	4	
	AZD2816	Wuhan-Hu-1	28 days after booster dose	GMT	5	[VmRNA:B2816:Wuhan]
				Seroresponse	6	
		B.1.351	28 days after booster dose	GMT	7	[VmRNA:B2816:Beta]
				Seroresponse	8	

GMT: Geometric mean titre

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 8 Immunogenicity Comparisons for Previously mRNA Vaccinated Group – Interim Analysis

Objective	$\frac{[\![\text{GMT}]\!]_{\text{comparator}}]}{[\![\text{GMT}]\!]_{\text{reference}}]}$	$[\![\text{Seroresponse}]\!]_{\text{comparator}}] - [\![\text{Seroresponse}]\!]_{\text{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD1222 booster dose in patients previously vaccinated with a mRNA vaccine is non-inferior to the response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{VmRNA: B1222: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Primary)	[VmRNA: B1222: Wuhan] − [HV1222(4): Wuhan: D2] (Other Secondary)
	$\frac{[\text{V1222: B1222: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Other Secondary)[a]	[VmRNA: B1222: Beta] − [V1222(4): Beta: D2] (Other Secondary)[a]
	$\frac{[\text{VmRNA: B1222: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Other Secondary)	[VmRNA: B1222: Beta] − [HV1222(4): Wuhan: D2] (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose in participants previously vaccinated with a mRNA is non-inferior to response elicited by a 2-dose AZD1222 vaccination	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.1)	[VmRNA: B2816: Beta] − [HV1222(4): Wuhan: D2] (Other Secondary)
	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{V1222(4): Beta: D2}]}$ (Key Secondary 2.5)[a]	[VmRNA: B2816: Beta] − [V1222(4): Beta: D2] (Other Secondary)[a]
	$\frac{[\text{VmRNA: B2816: Wuhan}]}{[\text{HV1222(4): Wuhan: D2}]}$ (Key Secondary 2.3)	[VmRNA: B2816: Wuhan] − [HV1222(4): Wuhan: D2] (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse elicited by an AZD2816 booster dose is non-inferior to response elicited by an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{VmRNA: B1222: Beta}]}$ (Key Secondary 2.2)	[VmRNA: B2816: Beta] − [VmRNA: B1222: Beta] (Other Secondary)
	$\frac{[\text{VmRNA: B2816: Wuhan}]}{[\text{VmRNA: B1222: Wuhan}]}$ (Key Secondary 2.4)	[VmRNA: B2816: Wuhan] − [VmRNA: B1222: Wuhan] (Other Secondary)

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

Table 8 Immunogenicity Comparisons for Previously mRNA Vaccinated Group – Interim Analysis

Objective	$\frac{[\![\text{GMT}]\!]_{\text{comparator}}]}{[\![\text{GMT}]\!]_{\text{reference}}]}$	$[\![\text{Seroresponse}]\!]_{\text{comparator}}] - [\![\text{Seroresponse}]\!]_{\text{reference}}]$
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD2816 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B2816: Beta}]}{[\text{VmRNA: B2816: Wuhan}]}$ (Other Secondary)	$[\text{VmRNA: B2816: Beta}] - [\text{VmRNA: B2816: Wuhan}]$ (Other Secondary)
To determine if the neutralizing antibody GMT response/seroresponse rate against the B.1.351 variant is non-inferior to the rate against the original Wuhan-Hu-1 strain following an AZD1222 booster dose in participants previously vaccinated with a mRNA vaccine	$\frac{[\text{VmRNA: B1222: Beta}]}{[\text{VmRNA: B1222: Wuhan}]}$ (Other Secondary)	$[\text{VmRNA: B1222: Beta}] - [\text{VmRNA: B1222: Wuhan}]$ (Other Secondary)

[a] As the immune response of AZD1222 against the B.1.351 variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses.

GMT: Geometric mean titre

Tables in grey are not planned to be performed at the interim but will be carried out during the primary analysis.

DocuSign Envelope ID: D2B7063B-D25E-4617-BF07-827D2D2C7286

6 REFERENCES

Little, R. J. A. and Rubin, D. B. Statistical Analysis with Missing Data, 2nd Edition, Hoboken, NJ: John Wiley & Sons 2002; 257.

Miettinen O, Nurminen M. Comparative analysis of two rates. Stat Med. 1985;4(2):213-26.

CHMP (Committee for Human Medicinal Products). Reflection paper on the regulatory requirements for vaccines intended to provide protection against variant strain(s) of SARS-CoV-2. https://www.ema.europa.eu/en/documents/scientific-guideline/reflection-paper-regulatory-requirements-vaccines-intended-provide-protection-against-variant_en.pdf. Published 2021.

Hing, J.P., Woolfrey, S.G., Greenslade, D. et al. Analysis of Toxicokinetic Data Using NONMEM: Impact of Quantification Limit and Replacement Strategies for Censored Data. J Pharmacokinet Pharmacodyn 28, 465–479 (2001). https://doi.org/10.1023/A:1012247131190

Due to technical issues, PPD (PPD) (ClinChoice) was not able to sign in ANGEL/CARA, but is signing instead in DocuSign.

DocuSigned by PPD
PPD
I am the author of this document
23 September 2022 | 18:39 GMT
8C9818B7DD7449E0A7174A550A2608ED

DocuSign

Certificate Of Completion

Envelope Id: D2B7063BD25E4617BF07827D2D2C7286
Status: Completed
Subject: Please DocuSign: d7220c00001-vmrna-sap-ed-2.0.docx
Source Envelope:
Document Pages: 32
Signatures: 1
Envelope Originator:
Certificate Pages: 3
Initials: 0
PPD
AutoNav: Enabled
EnvelopeId Stamping: Enabled
Time Zone: (UTC+05:30) Chennai, Kolkata, Mumbai, New Delhi
IP Address: 24.30.87.232

Record Tracking

Status: Original
9/23/2022 11:29:02 PM
Holder: PPD
Location: DocuSign

Signer Events	Signature	Timestamp
PPD PPD Security Level: Email, Account Authentication (Required)	PPD Signature Adoption: Pre-selected Style Signature ID: 8C9818B7-DD74-49E0-A717-4A550A2608ED Using IP Address: 70.63.230.6 With Signing Authentication via DocuSign password With Signing Reasons (on each tab): I am the author of this document	Sent: 9/23/2022 11:30:28 PM Viewed: 9/24/2022 12:07:43 AM Signed: 9/24/2022 12:10:00 AM
Electronic Record and Signature Disclosure: Accepted: 9/24/2022 12:07:43 AM ID: 966809fc-120e-46d2-a126-5466961d5d0e		

In Person Signer Events	Signature	Timestamp

Editor Delivery Events	Status	Timestamp

Agent Delivery Events	Status	Timestamp

Intermediary Delivery Events	Status	Timestamp

Certified Delivery Events	Status	Timestamp

Carbon Copy Events	Status	Timestamp

Witness Events	Signature	Timestamp

Notary Events	Signature	Timestamp

Envelope Summary Events	Status	Timestamps
Envelope Sent	Hashed/Encrypted	9/23/2022 11:30:28 PM
Certified Delivered	Security Checked	9/24/2022 12:07:43 AM
Signing Complete	Security Checked	9/24/2022 12:10:00 AM
Completed	Security Checked	9/24/2022 12:10:00 AM

Payment Events	Status	Timestamps

Electronic Record and Signature Disclosure

I hereby consent to that AstraZeneca Worldwide https://www.astrazeneca.com may disclose personal information such as; full name, email address, and any other information you may supply on the electronic form to AstraZeneca affiliates and third party service providers throughout the world in relation to the handling and administration of the Electronic Signature Service solution. This consent relates to any electronic records or signatures associated with the electronic contract.

AstraZeneca and the third party administering this service store and process personal information that AstraZeneca collects from you for the purposes of operating the Electronic Signature Service solution.

This also applies after termination of the Agreement. Processing of your personal information will be done in accordance with applicable law.

You may request access to your personal data and withdraw agreement to this processing at any time by contacting us in writing at docusign-support@astrazeneca.com.

Personal details and electronic signatures of signatories contained in contracts cannot be removed once the contract has been executed and will remain part of such contracts until these are destroyed in accordance with applicable law and AstraZeneca internal data retention policies.

SIGNATURE PAGE

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature

Document Name: d7220c00001-vmrna-sap-ed-2		
Document Title:	D7220C00001 VMRNA Statistical Analysis Plan Edition 2	
Document ID:	Doc ID-004787739	
Version Label:	2.0 CURRENT LATEST APPROVED	
Server Date **(dd-MMM-yyyy HH:mm 'UTC'Z)**	**Signed by**	**Meaning of Signature**
23-Sep-2022 20:47 UTC	PPD	Author Approval
23-Sep-2022 23:26 UTC	PPD	Content Approval

Notes: (1) Document details as stored in ANGEL, an AstraZeneca document management system.