EMA/351687/2022
Committee for Medicinal Products for Human Use (CHMP)

Type II variation assessment report

Procedure No. EMEA/H/C/005675/II/0052

Invented name: Vaxzevria

Common name: COVID 19 Vaccine (ChAdOx1 S [recombinant])

Marketing authorisation holder (MAH): AstraZeneca AB

Note

Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted and personal data anonymised.

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Status of this report and steps taken for the assessment			
Current step	**Description**	**Planned date**	**Actual Date**
☐	Start of procedure	24 Mar 2022	24 Mar 2022
☐	CHMP Rapporteur Assessment Report	06 Apr 2022	06 Apr 2022
☐	CHMP members comments	11 Apr 2022	11 Apr 2022
☐	Updated CHMP Rapporteur Assessment Report	13 Apr 2022	13 Apr 2022
☐	CHMP Rapporteur Assessment Report	04 May 2022	04 May 2022
☐	CHMP members comments	10 May 2022	10 May 2022
☐	Updated CHMP Rapporteur Assessment Report	12 May 2022	13 May 2022
☒	Opinion	19 May 2022	19 May 2022

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1. Background information on the procedure

Pursuant to Article 16 of Commission Regulation (EC) No 1234/2008, AstraZeneca AB submitted to the European Medicines Agency on 5 November 2021 an application for a variation.

The following changes were proposed:

Variation requested		Type	Annexes affected
C.I.4	C.I.4 - Change(s) in the SPC, Labelling or PL due to new quality, preclinical, clinical or pharmacovigilance data	Type II	I and IIIB

Update of sections 4.2, 4.8 and 5.1 of the SmPC in order to introduce a booster dose of Vaxzevria (homologous or heterologous) based on interim immunogenicity and safety data from the pivotal study D7220C00001, a partially double-blinded, randomised, multinational, active-controlled phase II/III clinical study and supportive literature evidence from studies COV001, RHH-001, COV-BOOST and Com-COV studies. The Package Leaflet is updated accordingly. In addition, the MAH took the opportunity to make minor editorial changes/corrections throughout the product information.

The requested variation proposed amendments to the Summary of Product Characteristics and Package Leaflet.

2. Introduction

Vaxzevria (also refer hereafter AZD1222) is indicated for active immunisation to prevent COVID-19 caused by SARS-CoV-2 in individuals 18 years of age and older. The approved posology consists of two separate doses of 0.5 mL each, to be administered intramuscularly. The second dose should be administered between 4 and 12 weeks (28 to 84 days) after the first dose.

The purpose of this variation is to support the use of AZD1222 as COVID-19 vaccination booster dose in adults 18 years and older, previously vaccinated with primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based). It presents a critical review of the benefits and risks of AZD1222 in this intended use, based on the clinical data summarised in the interim clinical study report (CSR) for Study D7220C00001 and supportive evidence from relevant clinical studies.

This assessment report (AR) summarises the available data on non-clinical, immunogenicity and safety data to support the above-mentioned variation. The product information has been updated accordingly.

3. Non-clinical aspects

3.1. Methods – analysis of data submitted

The MAH has provided additional non-clinical data in support of a third dose administration regarding pharmacology. Pharmacology data was submitted in the form of a peer reviewed publication by Spencer et al (Spencer et al 2021a, DOI: 10.1016/j.ebiom.2022.103902).

In the study BALB/c mice (n = 5 – 7/group) which have been previously vaccinated with 2 doses intramuscularly (IM) of AZD1222 4 weeks apart, were boosted 4 weeks later with a third dose of 10^8 infections units (iu) of either AZD1222 or AZD2816, a modified version of AZD1222 with the S glycoprotein gene from the Beta variant instead of the original Wuhan-Hu-1 strain. All mice were sacrificed 3 weeks post the third dose and antibody and T cell responses were assessed. It should be noted that the information provided also included data related to another product (Beta-AZD2816)

unrelated to the requested variation and therefore, this set of data is considered out of the scope of the evaluation.

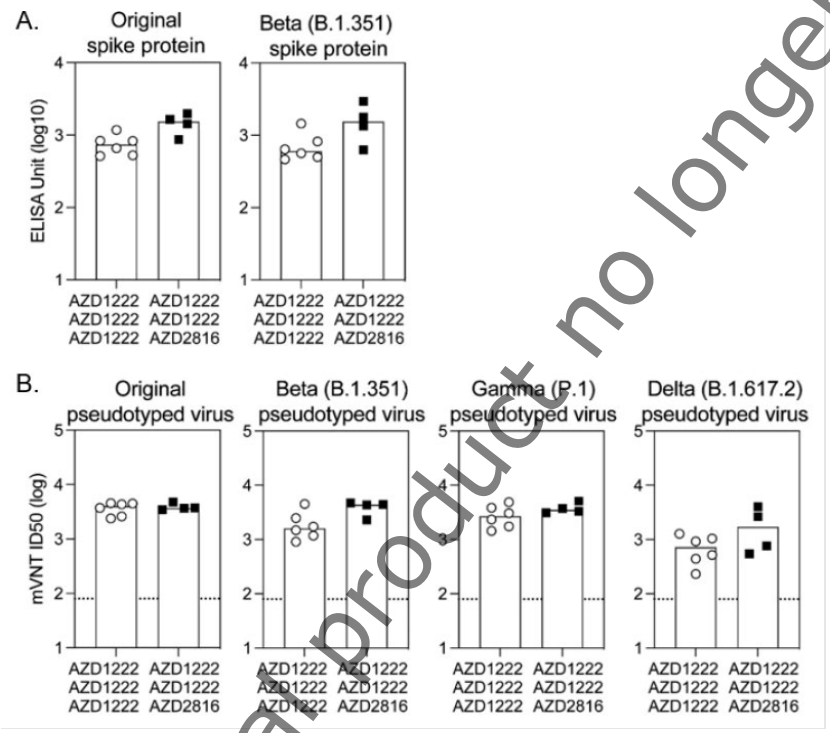
3.2. Results

An increase in spike-specific IgG was observed after the third dose of AZD1222 against all variants tested (wild-type and Beta spike protein; Figure 1 A), which was higher than the observed response following 2 doses.

Neutralising antibody responses were observed against wild-type, Beta, Delta and Gamma variants in a pseudovirus nAb assay (Figure 1 B) and were significantly higher when compared to 2 doses (Table 1).

As observed previously (Spencer et al 2021b), the anti-spike cell mediated response was primarily CD8+ T cells, with a high frequency of CD8+ T cells producing IFNγ and TNFα following a third dose of either AZD1222 or AZD2816 (Figure 2 C), with a majority of the response being T effector (Teff), T effector memory (Tem) CD8+ T cells.

Figure 1 Immune Response Following a Third Dose of AZD1222 or AZD2816



A) Total IgG level measured by ELISA against original spike protein (WT) or

B.1.351 spike protein. Data was log transformed and analysed with a two-way analysis of variance (repeated measure) and post-hoc positive test, no significance between groups ($p<0.05$) was observed.

B) Microneutralisation titres mVNT (ID80) measured against pseudotyped virus expressing original (WT),

B.1.351, B.1.617.2 or P.1 spike protein. Limit of detection in the assay is defined as a titre of 80 (dotted line). Data was log transformed and analysed with a two-way analysis of variance (repeated measure) and post-hoc positive test, no significance between groups ($p<0.05$) was observed.

Figure adapted from Spencer et al 2021a.

Table 1 Microneutralisation Titres Following a Boost of AZD1222

Prime	Boost	Boost	Time post last vaccine	Original wild-type spike		Beta (B.1.351)		Delta (B.1.617.2)		Gamma (P.1)	
				ID_{50}	ID_{80}	ID_{50}	ID_{80}	ID50	ID_{80}	ID_{50}	ID_{80}
AZD1222	AZD1222		20 days	1691 (613 to 2750	486 (134 to 712)	830 (729 to 1202)	243 (129 to 485)	151 (122 to 659)	93 (80 to 342)	2689 (1436 to 4861)	722 (162 to 1457)
AZD1222	AZD2816		20 days	2058 (1159 to 2815)	706 (477 to 926)	2281 (2000 to 4984)	585 (418 to 1371)	379 (158 to 827)	223 (85 to 454	3507 (3105 to 5100)	998 (714 to 2049)
AZD1222	AZD1222	AZD1222	20 days	4032 (2385 to 4559)	1478 (507 to 2222)	1609 (910 to 4519)	413 (240 to 1617)	721 (232 to 1274)	375 (123 to 745)	1896 (703 to 3610)	1017 (406 to 2322)
AZD1222	AZD1222	AZD2816	20 days	3704 (3462 to 4775)	2022 (1135 to 3949)	4392 (2304 to 4737)	1864 (767 to 2844)	1699 (547 to 4026)	615 (200 to 1816)	4755 (1637 to 5063)	3236 (1294 to 4968)

Prime	Boost	Boost	Time post last vaccine	Original wild-type spike		Beta (B.1.351)		Delta (B.1.617.2)		Gamma (P.1)	
				ID_{50}	ID_{80}	ID_{50}	ID_{80}	ID50	ID_{80}	ID_{50}	ID_{80}
AZD1222	AZD1222		20 days	1691 (613 to 2750	486 (134 to 712)	830 (729 to 1202)	243 (129 to 485)	151 (122 to 659)	93 (80 to 342)	2689 (1436 to 4861)	722 (162 to 1457)
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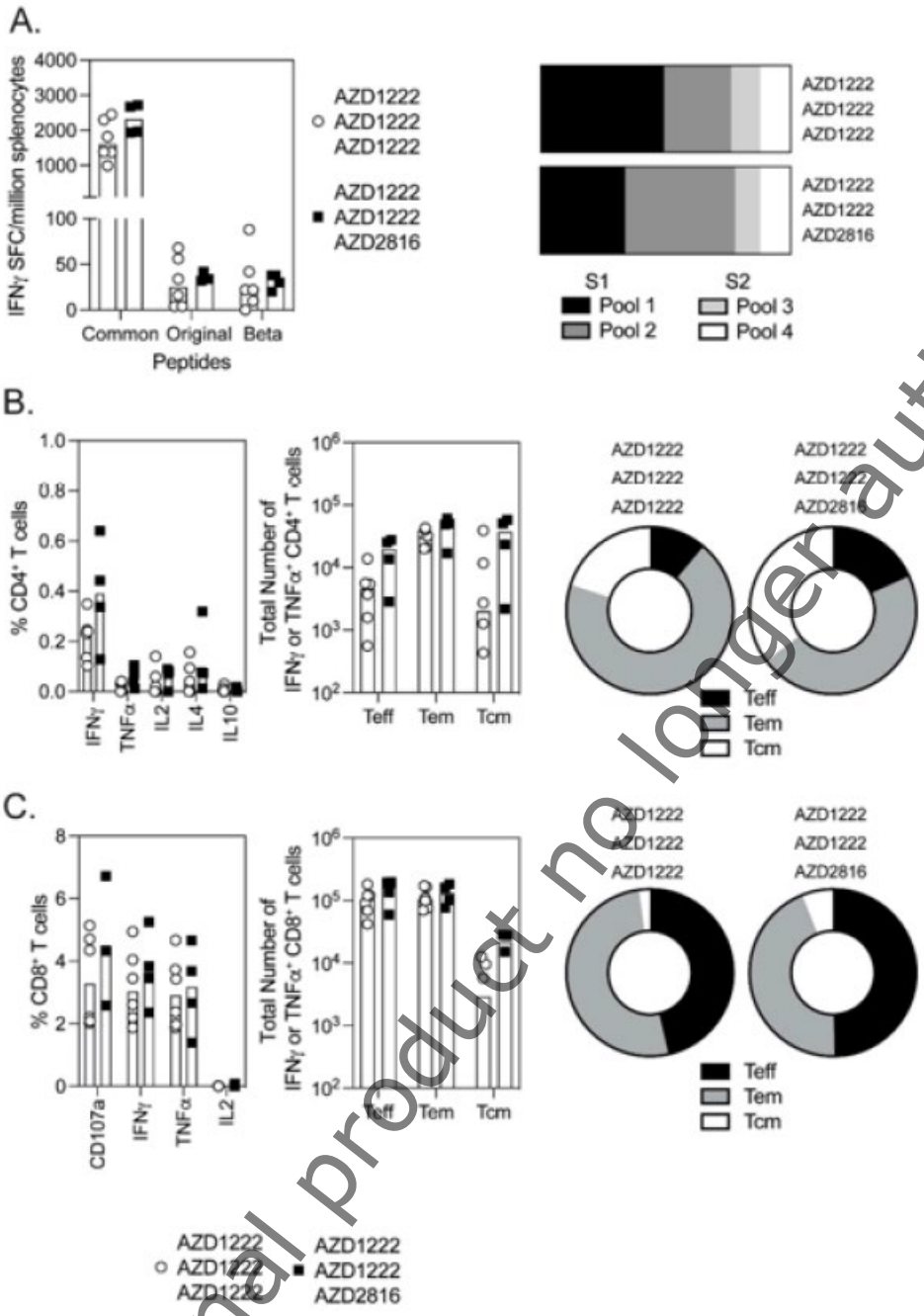
Functional ability of antibodies to neutralise pseudotyped virus expressing original spike, Beta (B.1.351), Delta (B.1.617.2) or Gamma (P.1) spike protein was measured in the serum of vaccinated mice. Pseudotyped virus neutralization titres are expressed as the reciprocal of the serum dilution that inhibited luciferase expression by 50% (ID_{50}) or 80% (ID_{80}). Table shows the median (min to max) per group.

Table adapted from Spencer et al 2021a.

Figure 2 T cell Responses Following Boost Vaccination



A. IFNγ secreting cells measured by ELISpot, with splenocytes stimulated with pools of common, original (WT) or B.1.351 peptides. Bar graph represents the proportion of IFNγ secreting cells measured against spike common peptides, sub-divided into S1 (pool 1 and pool 2) or S2 (pool 3 or pool 4) regions of spike protein.

B. Frequency of cytokine producing $CD4^+$, total number (left) or proportion (right) of $IFN\gamma^+$ or $TNF\alpha^+$ $CD4^+$ T cells of a T effector (Teff), T effector memory (Tem) or T central memory cells (Tcm) phenotype, bars represent the median response per group.

C. Frequency of cytokine producing $CD8^+$, total number (left) or proportion (right) of $IFN\gamma^+$ or $TNF\alpha^+$ $CD8^+$ T cells of a Teff, Tem or Tcm phenotype, bars represent the median response per group. Figure from Spencer et al 2021a.

3.3. Discussion

Data retrieved from a published paper has been submitted, in Balb/C mice in which immunogenicity resulting of administration of two (Prime boost) or three doses (boost dose) of AZD1222 (108 infectious units) has been assessed. Negative control animal groups have not been included in the study. Total IgG levels following immunization were measured against the original WT spike protein or

B.1.351. spike protein after reveal no significant differences between both settings, although a slightly improved response was seen against WT spike protein.

Neutralizing antibodies responses to immunization were also measured against pseudotyped virus expressing original (WT), B.1.351, B.1.617.2 or P.1 spike protein after 3 doses of AZD1222. Responses were generally higher in all instances as compared with two doses except for ID50 microneutralization titers observed against P.1 with has also shown a high degree of variability. The titers reported against WT original pseudotyped virus were the highest, followed by P.1, B.1.351 and B.1.617.2. The predominant cytokine response for both CD8+ and CD4+ T cells was expression of IFN-γ and TNF-α,

IFN-γ response was evaluated by ELISpot testing in splenocytes stimulated with peptide pools that were sub-divided into 4 pools to cover the S1 and S2 regions of spike. Data is indicative that the response was mainly driven by S1 (pools 1 and 2) which is consistent with previously submitted data after a prime boost regimen (2 doses). Relevant immune response for CD4+ and CD8+ T cell populations was also observed. Increases were notably higher in the CD8+ subtype compared to CD4+ in BALB/c mice. Most of the response reported was driven by T effector (Teff), T effector memory (Tem) CD8+ T cells. The leading cytokine response for CD8+ and CD4+ T cells was expression of IFN-γ and TNF-α. The administration of a booster dose did not result in a relevant increase of Tcell responses.

No additional endpoints of immune response have been submitted. This is considered acceptable from a non-clinical perspective since no relevant concerns have been identified in the data provided and the weight of immunogenicity boost response has been already addressed in the Clinical evaluation of this variation assessment.

4. Clinical Immunogenicity aspects

4.1. Methods – analysis of data submitted

Introduction

The purpose of this variation is to support the use of AZD1222 as COVID-19 vaccination booster dose in adults 18 years and older, previously vaccinated with primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based). It presents a critical review of the benefits and risks of AZD1222 in this intended use, based on the clinical data summarised in the interim CSR for Study D7220C00001 and supportive evidence from relevant clinical studies.

Study D7220C00001 is also designed to assess AZD2816 (a modified AZD1222 vaccine targeted against the Beta variant of SARS-CoV-2), as a third dose booster in previously vaccinated participants. Results to-date show that both AZD1222 and AZD2816 boosters elicited a robust immune response against all variants tested, with AZD2816 booster eliciting higher antibody titres against the Beta variant compared to the AZD1222 booster, as intended. No other remarkable differences with significant clinical implications, including safety, were noted between these two vaccines. Although both AZD2816 and AZD1222 demonstrated a positive risk-benefit profile as a third dose booster in Study D7220C00001, AZD2816 is not considered to offer sufficient differentiation from AZD1222 to warrant seeking an indication for the product at this time due its limited relevance in an epidemiological setting dominated by Delta and Omicron variants. Therefore, the clinical overview submitted by the MAH is primarily focused on data from the AZD1222 booster treatment arm and includes only a brief summary of results for AZD2816.

The MAH submitted an interim CSR that reports on an analysis of data from participants previously vaccinated against COVID-19 with 2 doses of either AZD1222 or an mRNA-based vaccine who then received a 1-dose booster of AZD2816 or AZD1222. Dosing of the previously unvaccinated cohort, who

are to receive a 2-dose primary series of AZD1222 and/or AZD2816, was still ongoing at the time of database lock for this interim analysis. The primary analysis, which includes comparative analyses across the previously vaccinated and previously unvaccinated cohorts, will occur once data for the previously unvaccinated cohort are available and will be reported in the Primary Analysis CSR.

Only descriptive immunogenicity results were originally planned to be presented in this interim CSR, with no comparative analyses. Following database lock both the MHRA and the CHMP requested that this interim analysis include comparative analyses, which were conducted against a matched cohort of participants from an historical study who had received 2 doses of AZD1222.

Although this is an interim analysis, it included a full analysis of the booster treatment group through Day 29 following AZD1222 booster. These clinical data include 689 participants, 30 years of age and older, including individuals with and without comorbidities that increase the risk for severe COVID-19. The majority of participants were seronegative at study start, but a small cohort of participants that were seropositive at baseline with previous SARS-CoV-2 infection are also included to support use across the real-world population.

Compliance with Good Clinical Practice

The sponsor's procedures, internal quality control measures and audit programmes provide reassurance that D7220C00001 was carried out in accordance with GCP, as documented by the ICH and applicable health authorities' guidelines.

Data Monitoring Committee

A COVID-19 Vaccine Data Safety Monitoring Board (DSMB) comprised of independent experts was convened to provide oversight and to ensure safe and ethical conduct of the study.

Regulatory Status

AZD1222 has a Conditional Marketing Authorisation in the EU and UK and is authorised in 93 countries worldwide.

In late 2021, AstraZeneca initiated global submissions to include administration instructions for a homologous boosting (third dose) with AZD1222 in the product label, based on the COV001 publication. To date, regulatory authorities have authorised an AZD1222 booster in 16 countries worldwide.

Conventions

This study includes multiple 'cohorts' and 'treatment groups'. The 2 main groups of participants, being (1) those previously vaccinated with AZD1222 or an mRNA vaccine and (2) those who were COVID-19 vaccine-naïve, are referred to as the 'previously vaccinated cohort' and the 'previously unvaccinated cohort', respectively. Within the previously vaccinated cohort there are 2 subcohorts, referred to as the 'AZD1222 cohort' (i.e., those who previously received 2 doses of AZD1222) and the 'mRNA cohort' (i.e., those who previously received 2 doses of a COVID-19 mRNA vaccine). There were 2 treatment groups (AZD1222 booster or AZD2816 booster) within each of these subcohorts.

Treatment groups are identified by 'V1222' and 'VmRNA', which refer to the cohorts based on their pre-study primary 2-dose course of vaccination, and by 'B1222' and 'B2816', which identify the booster dose received by that treatment group during the study. For example, V1222/B2816 refers to participants previously Vaccinated with 2 doses of AZD1222 who received a booster dose of AZD2816.

Regulatory History

Scientific advice related to Study D7220C00001 has been obtained as summarised in Table 2.

Table 2 Dates of Prior Scientific Advice

Topic	Date of health authority feedback		
	MHRA	EMA/CHMP	FDA
Clinical Trial Design	14 May 2021	21 May 2021	26 May 2021
Use of Historical Control	-	07 Oct 2021	-
Scientific Advice	29 Nov 2021	07 Dec 2021	-

CHMP, Committee for Medicinal Products for Human Use; CTA, clinical trial application; EMA, European Medical Agency; FDA, Food and Drug Administration; MAA, Marketing Authorisation Application; MHRA, Medicines and Healthcare Regulatory Authority

In general, the final design of the D7220C00001 study is aligned with the EMA, MHRA and FDA guidance (EMA guidance 2021, MHRA guidance 2021, FDA 2021) and feedback received from the Agencies. This included the non-inferiority analysis for GMT ratio and seroresponse rate, use of validated pseudovirus neutralisation assay, and inclusion of seropositive cohort to characterize reactogenicity and immunogenicity to AZD1222 and AZD2816.

AstraZeneca consulted with the CHMP on using historical controls as the primary vaccination comparators for the booster treatment arms instead of the in-study primary vaccination cohort, due to the urgency of the need for approved boosters. Based on unfeasibility to enrol unvaccinated participants older than 65 years old participants, CHMP agreed that the immunogenicity data on individuals of less than 65 years old could allow extrapolation to other age groups within the original indication.

In November 2021, AstraZeneca consulted with CHMP and MHRA on the adequacy of the clinical package based on the interim analysis of D7220C00001 study data to support an application for authorisation of AZD2816 as a 1-dose booster vaccination in individuals previously vaccinated against SARS-CoV-2 with AZD1222 or an mRNA vaccine, the use of alternate non-inferiority margin for GMT ratio and seroresponse difference, and to agree with the list of important prognostic covariates for primary analyses of GMT ratio using ANCOVA models. The CHMP responded by way of scientific advice provided on 07 December 2021 that key secondary endpoint 2.4 (i.e., comparing the response against the Wuhan-Hu-1 strain of a booster dose of AZD1222 to a 2-dose AZD1222 primary series) be promoted to a primary outcome. The CHMP also requested that the original SAP be separated into 3 individual SAPs specific to (1) the AZD1222 previously vaccinated cohort, (2) the mRNA previously vaccinated cohort, and (3) the previously unvaccinated cohort. These requests were implemented. The non-inferiority analysis of difference in seroresponse in this data has been conducted with the CHMP requested margin of -10%.

Following database lock on 17 November 2021, as per request from the MHRA and the CHMP, this analysis, which was originally planned to only present descriptive results for the previously vaccinated cohort, also includes comparative analysis to allow conclusive assessment of non-inferiority.

Overall Study Design of CT D7220C00001

Clinical trial D7220C00001 is an ongoing Phase II/III, partially double-blinded, randomised, multinational, active controlled study to evaluate the safety and immunogenicity of AZD2816 as a 1-dose booster vaccination in previously vaccinated adult participants and as a 2-dose primary vaccination in previously unvaccinated adult participants. This study is also investigating the safety and immunogenicity of (1) a 2-dose vaccination with AZD1222 as first dose and AZD2816 as the second dose and (2) a 1-dose booster of AZD1222 in participants previously vaccinated with a 2-dose COVID-19 vaccine.

A total of approximately 2590 SARS-CoV-2 nucleocapsid seronegative participants who were screened and judged to be eligible for the study were to be enrolled across these 2 populations. The goal was for 1300 previously vaccinated participants to receive an additional single-dose booster vaccination, and for

1290 unvaccinated participants to receive a 2-dose primary vaccination. In addition, seropositive participants were enrolled (with a maximum of 10% of the seronegative population or 259 participants) to support exploratory analyses in these participants.

The enrolment and randomisation strategy were intended to minimise group differences in terms of age, sex, and the presence of comorbidities.

The study contains 3 cohorts that were randomised to a total of 8 treatments:

- Approximately 700 seronegative participants who were previously vaccinated with 2 doses of AZD1222 were to be randomised 1:1 to 1 dose of AZD1222 or 1 dose of AZD2816.
- Approximately 600 seronegative participants who were previously vaccinated with 2 doses of an mRNA COVID-19 vaccine were to be randomised 1:1 to 1 dose of AZD1222 or AZD2816.
- Approximately 1290 seronegative, vaccination-naïve participants were to be randomized 5:5:5:2 to 2 doses of AZD1222 with a 4-week dosing interval, 2 doses of AZD2816 with a 4-week dosing interval, 1 dose of AZD1222 followed by 1 dose of AZD2816 with a 4-week dosing interval, or 2 doses of AZD2816 with a 12-week dosing interval.

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

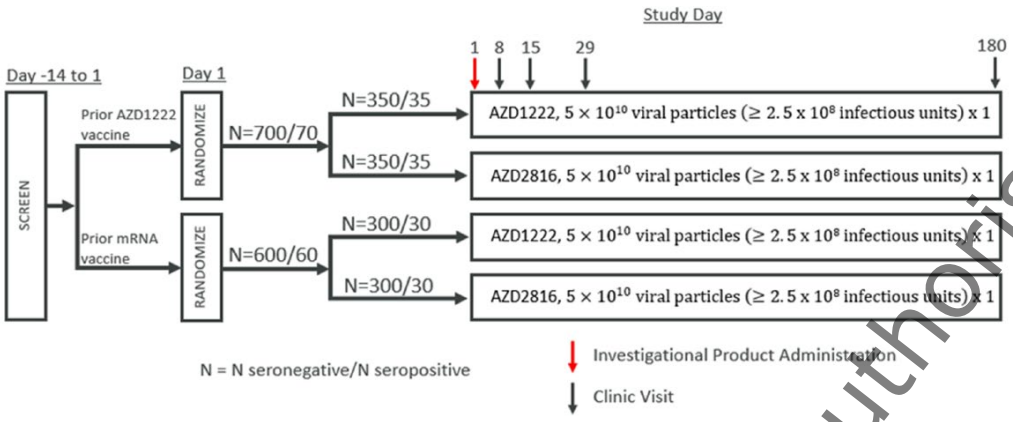
The 3 treatments with a 4-week dosing interval were double-blinded while the treatment with the 12-week interval was open-label due to the difference in dosing interval. The booster dose cohort was double-blinded.

Immunogenicity (i.e., anti-Wuhan-Hu-1 and anti-Beta immune responses including S-binding antibody titres and neutralising antibody levels [pseudoneutralisation]) were to 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. Responses to the Alpha and Gamma variants (S-binding antibody assay) and to the Delta variant (pseudoneutralisation assay) were also to be assessed. Peripheral blood mononuclear cells were to be isolated in a subgroup of participants to assess T- and B-cell responses.

All study participants were to be followed for safety for 180 days after administration of their last vaccination dose. In every participant, solicited local and systemic events were to 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, MAAEs, and AESIs were to be evaluated through study completion (up to 180 days after the last study vaccination).

Figure 3 shows the design of the study and the sequence of treatment periods for the previously vaccinated cohort.

Figure 3 Flow chart of Study design for previously vaccinated seronegative/seropositive participants receiving a 1-dose booster



Inclusion criteria

It follows a summary of the key inclusion criteria:

1. Adult, ≥ 18 years of age at the time of consent.

For inclusion in the SARS-CoV-2 seronegative population supporting the primary and secondary objectives:

2. No history of laboratory-confirmed SARS-CoV-2 infection (i.e., no positive nucleic acid amplification test and no positive antibody test).

3. Seronegative for SARS-CoV-2 at screening (lateral flow test to detect activity to the nucleoprotein).

Note, patients failing to meet criteria 2 and/or 3 may be included in the separate seropositive population supporting the seropositive exploratory objectives.

4. Medically stable, according to the judgment of the investigator.

5. Contraceptive use by women should be consistent with local regulations regarding the methods of contraception for those participating in clinical studies.

6. Prior completion of a 2-dose primary homologous vaccination regimen against the original SARS-CoV-2 Wuhan-Hu-1 strain with either AZD1222 (2 standard doses as authorised vaccine or as investigational product in a clinical trial with a 4- to 12-week dosing interval) or with an mRNA vaccine approved for emergency or conditional use (e.g., BNT162b2 vaccine [Pfizer-BioNTech] with a 3- to 12-week dosing interval or mRNA-1273 vaccine [Moderna] with a 4- to 12-week dosing interval). The second dose in all cases should have been administered at least 90 days prior to first administration of study intervention. Following a blinded review of protocol deviations related to the timing of doses, it was decided that only participants with intervals of less than 70 days from second dose to booster dose, or who had received their primary series of AZD1222 or an mRNA vaccine at an interval of less than 21 days or greater than 100 days, would be excluded from the immunogenicity analysis sets.

Exclusion criteria

It follows a summary of the key exclusion criteria:

1. History of allergy to any component of AZD1222/AZD2816.

2. History of Guillain-Barré syndrome, any demyelinating disease, or any other neuroimmunologic condition.

3. Significant infection or other acute illness, including fever > 100 °F (> 37.8 °C) on the day prior to or day of randomisation.

4. Any confirmed or suspected immunosuppressive or immunodeficient state, including asplenia or HIV/AIDS.

6. History of primary malignancy (some exceptions allowed)

7. Any other significant disease, disorder, or finding that may significantly increase the risk to the participant because of participation in the study, affect the ability of the participant to participate in the study, or impair interpretation of the study data.

8. Any autoimmune conditions, except mild psoriasis and vitiligo

Objectives and endpoints

Objectives

The initial primary and key secondary immunogenicity objectives for the previously vaccinated cohort, as specified in the CSP in effect at the time of database lock for this interim analysis, were as follows:

Primary:

1: To determine if the humoral immune 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.

Key secondary:

2.1: To determine if the humoral immune 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.

2.2: To determine if the humoral immune response elicited against the B.1.351 variant by an AZD2816 booster dose is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.

2.3: To determine if the humoral immune 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.

2.4: To determine if the humoral immune 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 a 2-dose AZD1222 vaccination.

2.5: To determine if the humoral immune response against the original Wuhan-Hu-1 strain elicited by an AZD2816 booster dose is non-inferior to the response elicited by an AZD1222 booster dose in participants previously vaccinated with AZD1222.

Following database lock, the CHMP requested changes to the Statistical Analysis Plan (see also previous section on "Regulatory History"). One of the requested changes was that the testing hierarchy for the immunogenicity endpoints be reordered, with key secondary endpoint 2.4 becoming the primary endpoint. The interim CSR submitted presents comparative analyses of immunogenicity results according to the hierarchy requested by the EMA. As requested by CHMP, the interim CSR submitted by the MAH include comparative analyses, conducted against a matched cohort of participants from an historical study who had received 2 doses of AZD1222. It is noted, that there were objectives (as for example Key secondary 2.1) where the comparator is the response elicited by 2-dose primary series AZD1222 vaccination against the Beta variant are not presented in this interim CSR since serum samples from historic control cohort participants were not tested against the Beta variant.

An additional request by the CHMP was that the primary analysis SAP be separated into 3 individual sub-SAPs specific to (1) the AZD1222 previously vaccinated cohort, (2) the mRNA previously vaccinated cohort, and (3) the previously unvaccinated cohort.

It follows the description of the objectives and endpoints of the two cohorts that received a booster dose of AZD1222:

AZD1222 vaccinated cohort that received a booster of AZD1222

According to the new specific SAP ("Previous AZD1222 Cohort Sub-SAP") the immunogenicity objective for the previous AZD1222 cohort that received a booster of AZD1222 were:

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:	
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 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	

Analysis of primary and secondary endpoints

The immunogenicity endpoints of interest in this study are:

- Geometric mean antibody titre of pseudoneutralising antibodies at Day 29
- Seroresponse, defined as ≥ 4-fold increase from baseline in pseudoneutralising antibodies at Day 29

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.

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. Analyses of GMT/GMR will be performed also on both unadjusted titre levels.

For GMT ratio, non-inferiority was 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) is > 0.67.

Regarding the secondary endpoint on the difference in seroresponse, non-inferiority was demonstrated if the lower bound of the 2- sided 95% CI rate difference in seroresponse between the comparator group and reference group was ≥-10%.

It is noted that 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.

Other secondary and exploratory endpoints

Additional immunogenicity endpoints included determination of immune response (in terms of GMT titers and seroresponse rates) using a pseudoneutralizing assays against a beta variant; spike-specific IgG response to SARS-CoV-2 to Wuhan strain VOCs beta, alpha and gamma by multiplexed immunoassay (S-protein binding antibody) and the anti-vector neutralizing antibody titres to the ChAdOx-1 adenovirus vector. For S-protein binding antibody results, since different assays were used between strains, and also within the same strains between this study and historical control study D8110C00001, these data will only be summarized descriptively, and no comparative analyses will be conducted.

The Delta variant analysis was based on an unvalidated pseudoneutralisation assay performed in a subpopulation of participants.

Another exploratory objective was to explore B-cell and T-cell responses following a booster dose of AZD1222 or AZD2816 in a subgroup of seronegative participants. The endpoint for this objective for the interim analysis was quantification of (IFN-γ) ELISpot responses to SARS-CoV-2 Beta or Wuhan-Hu-1 S protein over time.

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)

mRNA vaccinated cohort that received a booster of AZD1222

As detailed in the specific SAP (Previous mRNA Cohort Sub-SAP) for the mRNA vaccinated cohort that received an AZD1222 booster, the same primary/key secondary endpoints as described above for the AZD1222 cohort, are used. Moreover, the comparisons use the same comparator group of participants previously vaccinated with AZD1222 from a historical control cohort.

Thus, 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 an 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.

Similar analysis to those described above in section "Other secondary and exploratory endpoints" for the previously AZD1222 vaccinated cohort were also carried out for the previously mRNA vaccinated cohort.

Assays used to assess SARS-CoV-2 infection, immune response (humoral and cell-mediated), and anti-vector antibodies.

A listing of the immunogenicity bioanalytical methods used to obtain results for the primary, secondary and key exploratory objectives of the pivotal study is provided in Table 3. A tabular summary of immunogenicity assessments is provided in Table 4.

Table 3 Bioanalytical methods for assessment of primary, secondary and key exploratory endpoints in Study D7220C00001

Study	Protocol version	Assay	Endpoint Type	Antigen	Testing Laboratory	Validation Plan/Report
D7220C00001 (Phase II/III)	Version 1.0	PhenoSense pseudovirus SARS-CoV-2 nAb assay, pseudoneutralisation	Primary/ Secondary	Wuhan-Hu-1	Monogram Biosciences, South San Francisco, CA, USA	MG-SF-VALD-VR1038.000 PhenoSense Anti-SARS-CoV-2 nAb assay[a]
				B.1.351	Monogram Biosciences, South San Francisco, CA, USA	Bridging report pending[b]
		Multiplexed ECL Method for the Detection of SARS-CoV-2 S, N Antigens (5-Plex Variant of Concern)	Secondary	Wuhan-Hu-1, B.1.351, B.1.1.7, P.1, Nucleocapsid	PPD Laboratories, Richmond, VA, USA	Validation of A Multiplexed ECL Method for the Detection of SARS-CoV-2 Spike, SARS-CoV-2 Nucleocapsid, SARS-CoV-2 Spike P.1, SARS-CoV-2 Spike B.1.1.7, and SARS-CoV-2 Spike B.1.351 Antibodies in Human Serum (5-Plex Variant)[b]
		SARS-CoV-2 IFNγ ELISpot assay	Exploratory	Wuhan-Hu-1, B.1.351	University of Oxford, Oxford, UK	Validation of the ex-vivo IFNγ ELISPOT assay for use in clinical vaccine trials and immunology studies[c] Short method report: Analysis of the T cell response by T cell ELISpot assays to SARS-CoV-2 antigens[d]
		Anti-ChAdOx1 nAb assay	Exploratory	ChAdOx1	Monogram Biosciences, South San Francisco, CA, USA	MG-SF-VALD-VP1063.000 -FINAL[a]
		SARS-CoV-2 Delta Variant nAb assay	Exploratory	B.1.617.2	PPD Vaccines, Richmond, VA, USA	Not applicable

[a] Method validation report available on request.
[b] Will be submitted upon finalisation.
[c] For validation report, see report "Validation of the ex-vivo IFNγ ELISPOT assay for use in clinical vaccine trials and immunology studies", Module 5.3.1.4.
[d] For validation report, see report "Analysis of the T cell response by T cell ELISpot assays to SARS-CoV-2 antigens", Module 5.3.1.4.
nAb = neutralising antibody

Table 4 Immunogenicity and efficacy assessments in study D7220C00001

Study population age (years)	Group	Dose regimens evaluated	Dosing date (s)	Timing of second dose	Bioanalytical sampling dates
≥ 18	Previously Vaccinated and Naïve	NA	NA	NA	Blood sample for SARS-CoV-2 antibody testing (lateral flow test); Day -14 to Day 1
≥ 18	Previously Vaccinated	1 dose	D1	NA	Serum sample for SARS-CoV-2 serology testing: D1, D15, D29, D180
					Serum sample for exploratory assessment: D1, D15, D29, D180
					Serum sample for ChAdOx1 nAbs assessment: D1, D15, D29, D180
					Blood sample to assess B-cell and T-cell responses: D1, D15, D29, D180
					Blood sample for B-cell and T-cell response sequencing: D1, D15, D29, D180

It follows a brief description of the bioanalytical methods.

-PhenoSense Anti-SARS-COV-2 Pseudovirus Neutralising Antibody Assay (Wuhan-Hu-1 and B.1.351)

The PhenoSense Anti-SARS-CoV-nAb assay is based on previously described methodologies using HIV-1 pseudovirions (Petropoulos et al 2000, Richman et al 2003). The measurement of nAb activity using the PhenoSense SARS-CoV-2 nAb assay is performed by generating HIV-1 pseudovirions that express the SARS-CoV-2 Spike protein. Neutralising antibody activity is measured by assessing the inhibition of luciferase activity in HEK293 target cells expressing the ACE2 receptor, following pre-incubation of the pseudovirions with serial dilutions of the serum specimen. The expression of luciferase activity in target cells is inhibited in the presence of anti-SARS-CoV-2 nAb. Titres are reported as the ID50 of pseudovirus infection.

The method for the Wuhan-Hu-1 virus was performed at Monogram Biosciences in South San Francisco, CA, USA; the B.1.351 variant pseudovirus assay was also be performed at Monogram Biosciences. Validation for the Wuhan-Hu-1 virus included accuracy, repeatability, intermediate precision, linearity, specificity/selectivity, sensitivity and stability utilising pooled sera from high-titre, intermediate-titre and low-titre pooled convalescent SARS-CoV-2 sera, as well as historical negative samples collected in the year 2017 (prior to SARS-CoV-2 circulation). Validation included manual and automated sera dilution in either transiently transfected ACE-2 cells or stable ACE-2 cell lines. Validation to B.1.351 variant was completed with report signed off before the immunogenicity testing from study D7220C00001 began.

-Multiplexed ECL Method for the Detection of SARS-CoV-2 S, N Antigens (5-Plex Variant of Concern)

The indirect binding multiplexed ECL is a quantitative assay designed to detect antibodies to the SARS-CoV-2 in human serum. The assay is based on the MSD technology which employs multi-spot microtitre plates fitted with a series of electrodes associated with the bottom of each well. Antibody concentrations are determined in an indirect binding format. Specifically, the reference standard, quality control sample serum, and test samples are incubated on a MSD 96-well, 10-Spot Custom SARS-CoV2 Serology SECTOR® plate coated with SARS-CoV-2 S antigens for the Wuhan-Hu-1, B.1.351, B.1.1.7, and P.1 variants as well as the N antigen. To quantify the antigen response in AU/mL, a reference standard was created by pooling pre-screened COVID-19 positive human serum samples containing antibodies to S and N.

The multiplexed ECL method for the detection of antibodies to SARS-CoV-2 antigens was performed at PPD Vaccines laboratory in Richmond, VA, USA. The validation included measurements of precision and ruggedness, and to assess the dilutional linearity, selectivity and relative accuracy of the SARS-CoV-2 antigens Spike P.1, Spike B.1.1.7, and B.1.351. Additionally, the validation confirmed the previously established assay parameters for the S and N antigens from the reference SARS-CoV-2 wild-type strain. Assay validation was completed with reports signed-off before the immunogenicity testing from study D7220C00001 were performed.

-SARS-CoV-2 IFNγ ELISpot Assay

ELISpot assays were performed using cryopreserved PBMCs. The SARS-CoV-2 IFNγ ELISpot assay was performed at the University of Oxford (Oxford, UK).

Cells were plated at 250,000 cells per well and re-suspended to 5×10^6 cells/mL in R10 diluent. Synthetic peptides (15mers overlapping by 10 amino acids) were pooled and utilised to stimulate PBMCs; cells were incubated for 18 to 20 hours with pooled peptides. Responses are reported as mean SFC per million PBMCs by multiplying the average per stimulant by 4, with subtraction of the background count.

-Anti-ChAdOx1 Neutralising Antibody Assay

The Anti-ChAdOx1 nAb assay was performed at Monogram Biosciences in South San Francisco, CA, USA. The Anti-ChAdOx1 nAb assay utilises a recombinant chimpanzee adenovirus vector transduction assay to evaluate human serum for anti-ChAdOx1 nAb activity. Validation included valuation of accuracy, precision, linearity, linear range (ULOQ/LLOQ), specificity, selectivity and stability.

-SARS-CoV-2 Delta Variant Neutralising Antibody Assay (Unvalidated)

This was an exploratory assessment to test the nAb response to the Delta variant. This assay was developed for research purposes only and may be supplemented with data from a validated assay at a future data cut-off. The Delta variant MNA will be performed at PPD Vaccines (Richmond, VA, USA). The SARS-CoV-2 nAb assay for the Delta variant is a cell-based assay that is designed to determine the dilution (ID50) at which SARS-CoV-2 nAbs inhibit viral infection to 50% of the average virus control in 293T-ACE2 cells by Delta variant SARS-CoV-2 RVPs, which express green fluorescent protein. An HIV-1 based pseudovirus platform is utilised with a Delta (B.1.617.2) RVP containing mutations of: T19R, G142D, del156/157, R158G, L452R, T478K, P681R, D950N.

-Nab response to the SARS-COv-2 omicron variant.

In a separate CSR (study MS1222-0007), the anti-omicron antibodies present in subjects boosted with AZD1222, who were previously vaccinated with either AZD1222 or an mRNA vaccine previously, was analysed. The different assays were performed: the University of Oxford, Omicron live virus neutralisation (Oxford, UK) and the UKHSA Omicron Live virus neutralisation (Porton Down, UK).

Randomisation and Blinding

The randomised participants were centrally assigned to randomised study intervention using an Interactive Response Technology/Randomisation and Trial Supply Management.

Treatment was double-blinded for all previously vaccinated participants receiving a booster dose. For participants receiving double-blinded treatments, the randomisation code was not to be broken except in medical emergencies when the appropriate management of the participant required knowledge of the treatment randomisation.

Stratification

Randomisation was stratified based on age (< 65, ≥ 65), sex, and presence of at least one of the following comorbidities that are known risk factors for severe illness from COVID-19 (based on the participant's past and current medical history):

- Obesity (BMI ≥ 30 kg/m2 at baseline)
- Significant cardiovascular disease (eg, heart failure, coronary artery disease, congenital heart disease, cardiomyopathies, or pulmonary hypertension)
- Chronic lung disease (eg, chronic obstructive pulmonary disease, idiopathic pulmonary disease, cystic fibrosis, or moderate to severe asthma)
- Diabetes

Statistical Methods and Determination of Sample Size

An Interim SAP (iSAP; Edition 1 dated 17 November 2021) and a primary analysis SAP (Edition 3 dated 17 November 2021), outlining all planned analyses for the interim and primary analyses, respectively, were completed before unblinding of the data. After the interim data were unblinded to the study team, but before any data were provided to health authorities, both the MHRA and the CHMP requested changes to the planned analyses.

At the CHMP request the primary analysis SAP was converted into a Master SAP and 3 Sub-SAPs were created (V1222-SAP, VmRNA-SAP, and Naïve-SAP) all dated 01 February 2022 outlining descriptive and comparative immunogenicity analyses pertaining to administration of AZD1222 and AZD2816 to the 3 cohorts of enrolled participants: 1) previously vaccinated with AZD1222, 2) previously vaccinated with an mRNA vaccine, and 3) previously unvaccinated. The new V1222-SAP and VmRNA-SAP incorporate, and supersede, the iSAP.

Moreover, it was requested that the interim analysis included non-inferiority comparative analyses of immunogenicity. Comparative analyses in this interim CSR present the 95% confidence interval of both the GMT ratio and difference in seroresponse using non-inferiority margins of 1.5 and -10% respectively. In cases where the lower bound falls to the right of the non-inferiority margin, we infer the null hypothesis of inferiority can be rejected.

Geometric Mean Titres and Geometric Mean Fold Rise.

Geometric mean titres (GMT) and Geometric Mean Fold Rise (GMFRs) for antibody titres were calculated for each treatment received and summarised. For previously vaccinated study participants, the time point of interest was Day 29 post booster dose. For the historical control group, the primary time point of interest was Day 29 post dose 2 (Day 57). Descriptive statistics for GMTs and GMFRs include number of participants, geometric mean, 95% CI, minimum, and maximum.

The GMT was 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 was calculated as the antilogarithm transformation of the upper and lower limits for a two-sided CI for the mean of the log-transformed titres.

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

Seroresponse Rate

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

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

Non-Inferiority Testing.

Non-inferiority testing was conducted on GMT ratio and seroresponse rates based on the titre assessments for pseudoneutralising antibodies. The statistical methodology was based on a 2-sided 95% CI of the ratio of the GMTs or the difference in seroresponse rates, respectively.

For GMT ratio, non-inferiority was 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 is > 0.67.

For difference in seroresponse, non-inferiority was demonstrated if the lower bound of the 2-sided 95% CI rate difference in seroresponse between the comparator group and reference group was ≥-10%.

Immunogenicity Analysis Methodology.

For the previously vaccinated cohort, immunogenicity endpoints are summarised and analysed separately for the Seronegative Immunogenicity Analysis Set and the Seropositive Immunogenicity Analysis Set.

For this interim analysis, the GMT of antibody titre measurements and ELISpot results were evaluated 28 days after booster in participants previously vaccinated with AZD1222 or an mRNA vaccine. Analyses of GMT/GMR were performed on both unadjusted titre levels and ELISpot results, as well as on model-adjusted titre levels and ELISpot results. The model adjusted analyses were derived using analysis of covariance (ANCOVA) models that included the log transformed value of the titre or ELISpot result as the dependent variable, and independent variables for visit window (baseline, Day 15, Day 29), baseline co-morbidities (Yes or No), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, time since previous vaccination as a continuous (log-transformed) covariate, and participant as a random effect. The least square means for the visit effect and their 95% CIs were converted by anti-log into the adjusted GMT/GMR and its 95% CI at each visit.

Model fitting was performed within each treatment group. Model adjustments were performed for the Seronegative Immunogenicity Analysis Set only and were not performed on parameters with small sample sizes, in cases where the model does not converge nor has other performance issues. Model adjusted values use the mean time since previous vaccination across all previously vaccinated treatment arms.

Seroresponse rates were analysed using the same method as described earlier but using the model-adjusted baseline and post-baseline titre levels as described in the SAP. The model-adjusted titre levels were derived as the LS means for each treatment group and visit combination, plus the residual from the model fit. The LS means were obtained for a population with time since last vaccination set to the mean observed such time, balanced groups of Male and Female, comorbidity status, and age groups.

Historic Control Group

A matched historical control group was implemented for testing of the primary and certain secondary endpoints. Selection of the historic control group occurred before database lock and the unblinding of data.

This historical control group was matched to the cohort of participants previously vaccinated with a 2-dose primary series of AZD1222 based on age, gender, BMI, and presence of baseline comorbidities. These matched samples served as the control arm for all non-inferiority analyses (both GMT ratio and difference in seroresponse) of the previously vaccinated treatment groups to primary series vaccination.

A one-to-one propensity score matching was used to match seronegative historical control participants from study D8110C00001 in the immunogenicity analysis set who were previously vaccinated by a 2-dose AZD1222 vaccination (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 (referred to as 'cases').

A list of controls was provided based on adjustment of gender, age, BMI, and presence of baseline comorbidities. The selection of controls using these covariates was based upon blinded data from this study, 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. This was done using backwards selection when fitting the logistic regression on study membership.

Once the covariates were selected, a one-to-one full matching algorithm was used to provide the list of matched controls from study D8110C00001. Exact matching was done with respect to factor variables. A caliper was considered to ensure a common support region is established for the covariates from each study. The distance metric between matched controls and participants from this study was defined based on the Mahalanobis distance derived from the logit of the propensity score adjusting for BMI and age. 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.

Based on this list of controls the subset of participants who received both primary vaccinations of AZD1222, had pseudoneutralising antibody assessments at baseline and 29 days post dose 2, and prior to 29 days post dose 2 had no SARS-CoV-2 infection during the study and did not receive any immune-modifying drugs, blood products, or vaccines, were selected for the analysis. See Table 11 for matching criteria summary data from the historical control and AZD1222 previously vaccinated cohorts (shown later in this report).

Comparisons of antibody titres between the previously vaccinated cohort in this study and the historical controls from Study D8110C00001 were conducted using the Seronegative Immunogenicity Analysis Set, on the subset of historical control participants who had pseudoneutralising titre assessments at both baseline and Day 29 post dose 2 (i.e., using an adjusted ANCOVA model similar to the model adjustment described above, without the inclusion of a term for time since previous vaccination to calculated adjusted means and standard errors for the historical comparators, and also on unadjusted titres). Study D8110C00001 utilised the same validated pseudovirus neutralising antibody assay for the Wuhan-Hu-1 strain as was used for this study.

Methods for Multiplicity Control

A hierarchical approach was used to control for multiplicity of the primary and key secondary immunogenicity endpoints. That is, the null hypothesis for the immunogenicity endpoints was tested in a hierarchical order, and the subsequent null hypothesis was tested only if the prior null hypothesis was rejected. Consequently, no adjustment to alpha for multiplicity was made in the analysis of immune response. Separate hierarchies are used for participants previously vaccinated with AZD1222, and participants previously vaccinated with a mRNA vaccine for the interim, with separate type I error rate controls.

All summaries and analyses utilise the 95% confidence interval (type I error rate of 5%).

Description of Analysis Sets

The analysis sets are defined in the next table:

Table 5 Analysis Sets

Population/Analysis Set	Description
All Participants Analysis Set	All participants screened for the study, to be used for reporting disposition and screening failures.

Population/Analysis Set	Description
Full Analysis Set	All randomised participants who received study treatment, irrespective of their protocol adherence and continued participation in the study. Participants were analysed according to their randomised treatment, irrespective of whether or not they prematurely discontinued, according to the intent-to-treat principle. Participants who withdrew consent or assent to participate in the study were included up to the date of their study termination.
Safety Analysis Set	The Safety Analysis Set consists of all participants who received study treatment. Erroneously-treated participants (eg, those randomised to AZD2816, but were actually given treatment AZD1222) were accounted for in this analysis set by assigning them to the treatment they actually received.
Immunogenicity Analysis Set	The vaccine Immunogenicity Analysis Set includes all randomised participants who received at least 1 dose of planned study treatment (ie, 1 dose of either AZD1222 or AZD2816), had baseline and post-dose antibody measurements, had 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 were based on the actual treatment received.
Seronegative Safety Analysis Set	The subset of Safety Analysis Set participants who were seronegative at baseline.
Seropositive Safety Analysis Set	The subset of Safety Analysis Set participants who were seropositive at baseline.
Seronegative Immunogenicity Analysis Set	The subset of Immunogenicity Analysis Set participants who were seronegative at baseline.
Seropositive Immunogenicity Analysis Set	The subset of Immunogenicity Analysis Set participants who were seropositive at baseline.

Determination of Sample Size

Enrolment in this study was intended to result in randomisation of 2590 SARS-CoV-2 nucleocapsid seronegative participants: 1290 previously unvaccinated and 1300 previously vaccinated. Approximately 700 participants previously vaccinated with AZD1222 and approximately 600 participants previously vaccinated with an approved mRNA-based vaccination were to be randomised 1:1 to receive a single dose of AZD1222 or AZD2816. In addition, seropositive participants were to be enrolled (with a maximum of 10% of the seronegative population or 259 participants) to support exploratory analyses in these participants.

If there is no difference between treatment arm of interest (i.e., a ratio of 1) in the proportion of seroresponders, 380 participants provided 98% power to establish non-inferiority to within a margin of -15%, and 79% power to establish non-inferiority to within a margin of -10%, if the seroresponse rate is > 50%. The observed pseudoneutralising response rates (≥ 4-fold increase from baseline) from the COV001/002/003/005 studies for AZD1222 were 59.7% and 85.5% for the 4-week and 12-week dosing interval respectively (. A population of 380 participants provides 99% power to detect non-inferiority using a margin of -15%, and 81% power to detect non-inferiority using a margin of -10%, if the observed response rate is 59.7%.

Interim Analyses

A pre-specified initial interim analysis was performed on a subset of previously AZD1222 vaccinated participants that had received a booster dose to consider unblinded sample size adjustment. Access to the results of this interim analysis was restricted to an unblinded team that was not involved in the ongoing operation or reporting of the continuing clinical study.

This interim CSR presents the results of the pre-specified second interim analysis, performed once all previously AZD1222-vaccinated participants had completed their Day 29 visit. Additionally, the comparative analyses requested by the CHMP and MHRA are presented.

The final analysis will occur when data from all vaccinated participants is available through completion of the last study visit at 180 days after the final dose of study intervention.

Changes in the Conduct of the Study or Planned Analyses

Clinical Study Protocol Amendments

Unless stated otherwise, all references to the CSP refer to the version in effect at the time of interim analysis database lock (i.e., Amendment 3 dated 11 October 2021).

The original CSP was dated 14 May 2021. Changes in the conduct of the study that were implemented by protocol amendment 1 (02 June 2021), 2 (29 July 2021), and 3 (11 October 2021), as well as a local UK amendment regarding the minimum age of UK study participants.

Table 6 Protocol amendments related to changes in study conduct

Amendment Number/Date	Key details of amendment	Main reason(s) for amendment
Amendments made *before* the start of participant recruitment		
Amendment 1 (02 June 2021)	Addition of 2 treatment arms: 1) AZD1222 as a single booster vaccination in participants previously vaccinated with an mRNA COVID-19 vaccine and 2) heterologous vaccination with AZD1222 plus AZD2816 in previously unvaccinated participants. Further definition of analysis sets. Addition of thrombotic events with thrombocytopenia as a discontinuation criteria.	To incorporate feedback from internal and regulatory authority reviews
Local Amendment GBR-1 (UK; 03 June 2021)	Restrict the UK study population to adults ages 30 and above and provide a risk:benefit statement for the inclusion of adults ages 30 to 39	To incorporate feedback from MHRA
Amendments made *after* the start of participant recruitment		
Amendment 2 (29 July 2021) and Local Amendment	Added an additional interim analysis to evaluate immunogenicity in a subset of AZD1222 previously vaccinated subjects boosted with AZD1222 or AZD2816	To allow the early review of data on booster doses in the context of the ongoing pandemic
GBR-2 (UK; 30 July 2021)	Revised Objectives/Endpoints from descriptive to comparative, with ranking of primary, key secondary, other secondary, and exploratory objectives	Comparative analyses were added to show non-inferiority across treatments and allow for the submission of data from this study to support health authority approvals.
	Added non-inferiority margins to primary analysis and add additional participants to maintain power	To support comparative analyses.
Amendment 3 (11 October 2021) and Local Amendment GBR-3 (UK; 12 October 2021)	Removed the age cap regarding the previously unvaccinated cohort	Due to difficulties in recruiting elderly participants to the previously unvaccinated cohort
	Revised the primary and key secondary non-inferiority analyses of the previously vaccinated cohort to include historical controls, and include the statistical approach to be used	Inclusion of historic controls required based on anticipated confounding between previously vaccinated and previously unvaccinated cohorts.

MHRA, Medicines and Healthcare Products Regulatory Agency; UK, United Kingdom

Changes to Planned Analyses

As detailed above, the control arm was originally planned to be the 2-dose AZD1222 treatment group in this study, but the recruitable primary vaccination population was found to have few participants ≥ 65 years of age and was unable to meet the 25% minimum quota for that group. Given that a large difference in the mean age between the booster and primary vaccination cohorts of participants may confound comparisons between treatment arms across cohorts, it was determined that a matched historic control group of participants from study D8110C00001, conducted in the US and Latin America, would provide the most robust comparison. Following positive feedback from the CHMP, the CSP was amended on 11 October 2021 to incorporate this change. Selection of the historic control group occurred before database lock and the unblinding of data.

Following database lock on 17 November 2021, both the MHRA and the CHMP requested that this interim analysis, which was originally planned to only present descriptive results for the previously vaccinated cohort, also include comparative analyses. These requests were made after the interim data were unblinded to the study team but before any data were provided to the MHRA or the EMA. Comparative analyses based on the interim analysis dataset were subsequently conducted and are presented in this Interim CSR, these analyses are not considered to constitute the formal analyses of the primary and key secondary objectives. While these analyses will be updated during the primary analysis there were complete data available for pseudoneutralising antibodies at interim analysis database lock and, as such, the primary and key secondary results are not anticipated to change when using the primary analysis dataset. Therefore, the data available at this interim analysis allowed for hypothesis testing and conclusive assessment of the primary and most key secondary endpoints.

The CHMP also requested, by way of scientific advice provided on 07 December 2021, that key secondary endpoint 2.4 (ie, comparing the response against the Wuhan-Hu-1 strain of a booster dose of AZD1222 to a 2-dose AZD1222 primary series) be promoted to a primary outcome on the basis that this was the most relevant objective in supporting the use of AZD1222 as a booster dose. The CHMP also requested that the original primary analysis SAP be separated into 3 individual SAPs specific to (1) the AZD1222 previously vaccinated cohort, (2) the mRNA previously vaccinated cohort, and (3) the previously unvaccinated cohort. Lastly, the CHMP requested that non-inferiority conclusions from comparative analyses of seroresponse be based upon a margin of -10% rather than the -15% used for sample sizing in the study protocol. These requests were implemented. For the revised testing hierarchy for the AZD1222 cohort, and the equivalent testing hierarchy for the mRNA cohort, see Appendix 16.1.9 for the SAPs dated 01 February 2022.

As the immune response of AZD1222 against the Beta variant was not assessed in study D8110C00001, the AZD1222 treatment group from the previously unvaccinated cohort will be utilised for affected comparative analyses. The results of this and other comparisons where control group response to the Beta variant is the comparator will be reported in the Primary Analysis CSR.

Interim Analyses

This Interim CSR presents the results of the prespecified second interim analysis, performed once all previously AZD1222-vaccinated participants had completed their Day 29 visit. Additionally, the comparative analyses requested by the CHMP and MHRA are presented.

The final analysis will occur when data from all vaccinated participants is available through completion of the last study visit at 180 days after the final dose of study intervention.

4.2. Results

Study participants

The data presented in this section are limited to the seronegative cohort unless otherwise specified.

This study is being conducted at 35 sites in Brazil, Poland, South Africa, and the UK. Previously vaccinated participants from 19 study sites in the UK and 4 study sites in Poland contributed to this interim analysis. The first participant was enrolled on 27 June 2021. The data cut-off date for the results presented in this Interim CSR was 11 October 2021 and database lock occurred on 17 November 2021. At interim analysis database lock, data through Day 29 were available for all seronegative previously vaccinated participants but not for all seropositive previously vaccinated participants; only participants with data available through Day 29 (or who died or withdrew from the study before Day 29) were included in the interim analysis dataset and are discussed below.

In total, 1581 participants were screened, 1380 previously vaccinated participants were randomised, and 1379 of these participants received a booster dose of AZD2816 or AZD1222. The last participant randomised to this cohort received the booster dose on 10 September 2021. The most common reason for not being randomised was failure to meet the inclusion/exclusion criteria. At the time of the data cut-off on 11 October 2021, 1376 (99.8%) participants were continuing in the study.

Figure 4 summarises the disposition for all previously vaccinated participants (i.e., both seronegative and seropositive). The disposition of participants was generally balanced across treatment groups.

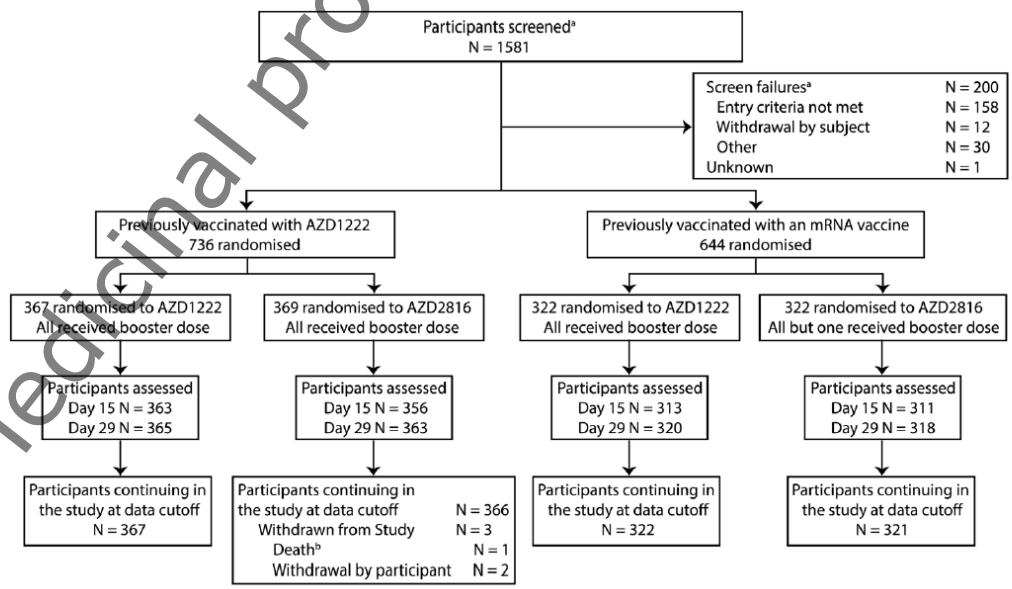
Only 3 participants were withdrawn from the study at the time of data cut-off, all from the V1222/B2816 treatment group (1 death and 2 participant decisions).

Of the 644 randomised participants in the mRNA cohort, all but 1 had been previously vaccinated with the BNT162b2 vaccine. The remaining participant had received mRNA-1273. This is reflective of the timing of vaccine approvals and rollout in the countries where this study is being conducted.

Data for the previously vaccinated cohort that were not available at the time of interim analysis database lock but will appear in the primary analysis dataset include:

- Spike-specific IgG response to SARS-CoV-2 by multiplexed immunoassay (S-protein binding antibody) samples for approximately 26 participants.
- Day 29 data for approximately 17 seropositive participants, owing to their time of enrolment.

Figure 4 Participant disposition and study participation – previously vaccinated cohort



[a] Participants screened and screen failures includes unvaccinated participants. These participants may have been eligible to participate in the 2-dose primary series cohort but were considered screen failures for the purposes of the previously vaccinated cohort.
[b] The death, due to pancreatic adenocarcinoma, was reported as not related to study intervention (Section 12.3.1).
Derived from: Table 14.1.1

Protocol Deviations

Protocol deviations occurred in a similar proportion of study participants across all 4 treatment groups (see Table 7).

Most of the deviations categorised as 'important protocol deviations' were related to, e.g., study visits occurring outside of planned visit windows, missing e-diary entries, missing laboratory data, or incorrect stratification at randomisation. There were only 9 (0.7%) participants with protocol deviations that were deemed to interfere with the generation or interpretation of an immune response. These deviations included receipt of study intervention < 70 days post-second dose of primary series vaccination and having severe liver disease. Few (n = 10) protocol deviations were related to COVID-19.

Table 7 Important protocol deviations (Full analysis set)

Subjects with at least one important protocol deviation interfering with the generation or interpretation of an immune response	3 (0.8)	0	3 (0.9)	3 (0.9)	9 (0.7)

	V1222 B1222 N = 367 n (%)	V1222 B2816 N = 369 n (%)	VmRNA B1222 N = 342 n (%)	VmRNA B2816 N = 321 n (%)	Total N = 1379 n (%)
Subjects with at least one important protocol deviation	161 (43.9)	166 (45.0)	146 (45.3)	147 (45.8)	620 (45.0)
Administrative Criteria	70 (19.1)	80 (21.7)	63 (19.6)	62 (19.3)	275 (19.9)
Eligibility and Entry Criteria	48 (13.1)	45 (12.2)	28 (8.7)	33 (10.3)	154 (11.2)
Informed Consent	0	0	1 (0.3)	2 (0.6)	3 (0.2)
IP Compliance	0	0	0	1 (0.3)	1 (0.1)
Laboratory Assessment Criteria	69 (18.8)	60 (16.3)	42 (13.0)	40 (12.5)	211 (15.3)
Randomisation Criteria	2 (0.5)	3 (0.8)	4 (1.2)	2 (0.6)	11 (0.8)
Study Procedures Criteria	63 (17.2)	64 (17.3)	56 (17.4)	51 (15.9)	234 (17.0)
Visit Schedule Criteria	28 (7.6)	32 (8.7)	30 (9.3)	31 (9.7)	121 (8.8)
Subjects with at least one important protocol deviation interfering with the generation or interpretation of an immune response	3 (0.8)	0	3 (0.9)	3 (0.9)	9 (0.7)
Eligibility and Entry Criteria	3 (0.8)	0	3 (0.9)	3 (0.9)	9 (0.7)
Subjects with at least one important protocol deviation related to COVID-19	2 (0.5)	2 (0.5)	3 (0.9)	3 (0.9)	10 (0.7)
Eligibility and Entry Criteria	0	1 (0.3)	1 (0.3)	1 (0.3)	3 (0.2)
Study Procedures Criteria	1 (0.3)	0	2 (0.6)	1 (0.3)	4 (0.3)
Visit Schedule Criteria	1 (0.3)	1 (0.3)	0	1 (0.3)	3 (0.2)
Subjects with at least one important protocol deviation excluding COVID-19 related deviations	160 (43.6)	166 (45.0)	145 (45.0)	146 (45.5)	617 (44.7)
Administrative Criteria	70 (19.1)	80 (21.7)	63 (19.6)	62 (19.3)	275 (19.9)
Eligibility and Entry Criteria	48 (13.1)	45 (12.2)	28 (8.7)	33 (10.3)	154 (11.2)
Informed Consent	0	0	1 (0.3)	2 (0.6)	3 (0.2)
IP Compliance	0	0	0	1 (0.3)	1 (0.1)
Laboratory Assessment Criteria	69 (18.8)	60 (16.3)	42 (13.0)	40 (12.5)	211 (15.3)
Randomisation Criteria	2 (0.5)	3 (0.8)	4 (1.2)	2 (0.6)	11 (0.8)
Study Procedures Criteria	62 (16.9)	64 (17.3)	54 (16.8)	50 (15.6)	230 (16.7)
Visit Schedule Criteria	27 (7.4)	32 (8.7)	30 (9.3)	30 (9.3)	119 (8.6)

Percentages are based on N, the number of subjects in the analysis set for each treatment group.
The same subject may have more than one important protocol deviation.
n Number of subjects per category.
V primary vaccination; B booster vaccination.

Study Participants Analysed (Analysis Sets)

Table 8 summarises the analysis sets and the number of participants in each analysis set for the previously vaccinated cohort.

Data presented in this Interim CSR are for the Seronegative and Seropositive Immunogenicity Analysis Sets.

Table 8 Analysis Sets- Previously vaccinated cohort

	Number of Participants				
	V1222/ B1222	V1222/ B2816	VmRNA/ B1222	VmRNA/ B2816	Total
Participants randomised	367	369	322	322	1380
Did not receive study intervention	0	0	0	1[a]	1[a]
Full Analysis Set[b]	367	369	322	321	1379
Safety Analysis Set	367	368[c]	322	322[c]	1379
Seronegative Safety Analysis Set[d]	347	349	299	301	1296
Seropositive Safety Analysis Set[e]	20	19	23	21	83
Immunogenicity Analysis Set[f]	362	360	317	315	1354
Excluded from the analysis set[g]	5	8	5	7	25
Did not have baseline and post-dose antibody measurement	3	8	3	5	19
Did not have at least one post-dose quantifiable serum titre	1	4	2	2	9
Had protocol deviations judged to have the potential to interfere with the generation or interpretation of an antibody response	2	0	2	2	6
Seronegative Immunogenicity Set[f]	342	341	294	294	1271
Seropositive Immunogenicity Set[f]	20	19	23	21	83

[a] This participant withdrew consent the same day as randomisation but before the booster dose was administered (see Listing 16.2.1.1).

[b] Participants randomised who received study intervention, irrespective of their protocol adherence and continued participation in the study. Participants analysed according to their randomised study intervention assignment, irrespective of the study intervention actually received.

[c] One participant randomised to the V1222/B2816 treatment group had previously received an mRNA vaccine, not AZD1222. This participant is included in the V1222/B2816 treatment group for the Full Analysis Set but in the VmRNA/B2816 treatment group for the Safety and Immunogenicity Analysis Sets.

[d] Seronegative participants who received at least 1 dose of study intervention. Participants analysed according to study intervention actually received.

[e] Seropositive participants who received at least 1 dose of study intervention. Participants analysed according to study intervention actually received.

[f] Immunogenicity analysis sets include all randomised participants who received study intervention, had baseline and post-dose antibody measurements, had 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. Participants analysed according to study intervention actually received.

[g] Count of participants included in the Safety Analysis Set but excluded from the Immunogenicity Analysis Set. An individual participant could have been excluded for more than 1 reason.

Demographic and Other Participant Characteristics

Table 9 summarises the demographic characteristics of study participants in the Seronegative Immunogenicity Analysis Set. Almost all (98.4%) of the previously vaccinated cohort was randomised at sites in the UK and most (88.3%) were White. The study population was generally representative of the previously vaccinated UK population at the time of enrolment.

Table 9 Demographic characteristics (Seronegative Immunogenicity Analysis Set)

Characteristic/Statistic	V1222/ B1222 N = 342	V1222/ B2816 N = 341	VmRNA/ B1222 N = 294	VmRNA/ B2816 N = 294	Total N = 1271
Age (years), n [a]					
Mean	59.7	60.5	55.5	56.1	58.1
SD	13.6	13.2	13.2	13.7	13.6
Median	62.0	62.0	55.0	55.0	58.0
Min	30	30	30	30	30
Age group (years), n (%) [a]					
≥ 18 to < 65	185 (54.1)	184 (54.0)	215 (73.1)	216 (73.5)	800 (62.9)
≥ 65	157 (45.9)	157 (46.0)	79 (26.9)	78 (26.5)	471 (37.1)
Sex, n (%)					
Male	186 (54.4)	184 (54.0)	112 (38.1)	116 (39.5)	598 (47.0)
Female	156 (45.6)	157 (46.0)	182 (61.9)	178 (60.5)	673 (53.0)
Race, n (%)					
White	298 (87.1)	296 (86.8)	265 (90.1)	263 (89.5)	1122 (88.3)
Black or African American	2 (0.6)	1 (0.3)	3 (1.0)	2 (0.7)	8 (0.6)
Asian	9 (2.6)	12 (3.5)	8 (2.7)	11 (3.7)	40 (3.1)
Mixed	0	1 (0.3)	2 (0.7)	0	3 (0.2)
Unknown	33 (9.6)	31 (9.1)	16 (5.4)	18 (6.1)	98 (7.7)
Ethnic group, n (%)					
Hispanic or Latino	6 (1.8)	9 (2.6)	3 (1.0)	6 (2.0)	24 (1.9)
Not Hispanic or Latino	295 (86.3)	300 (88.0)	268 (91.2)	265 (90.1)	1128 (88.7)
Missing	41 (12.0)	32 (9.4)	23 (7.8)	23 (7.8)	119 (9.4)
Country, n (%)					
Poland	10 (2.9)	6 (1.8)	1 (0.3)	3 (1.0)	20 (1.6)
United Kingdom	332 (97.1)	335 (98.2)	293 (99.7)	291 (99.0)	1251 (98.4)

[a] Age at randomisation.
Percentages are based on N, the number of subjects in the analysis set for each treatment group.
B1222, Participants receiving a third dose booster of AZD1222; B2816, Participants receiving a third dose booster of AZD2816; Max, Maximum; Min, Minimum; N, Number of participants in treatment group; SD, Standard deviation; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222.

Table 10 summarises the baseline characteristics of study participants in the Seronegative Immunogenicity Analysis Set. Similar to the demographic characteristics, there were differences between the AZD1222 and mRNA cohorts.

There was to be a minimum of 90 days between previous vaccination and booster dose. There were 4 previously vaccinated seronegative participants who received the booster dose < 70 days after their second dose. These participants were excluded from the Seronegative Immunogenicity Analysis Set. There were 13 seronegative participants who received the booster dose ≥ 70 days but < 90 days after their second dose, 4 participants were in the V1222/B1222 treatment group and 5 participants were in the VmRNA/B1222 treatment group, and 4 participants were in the VmRNA/B2816 treatment group. It was decided, prior to database lock, to include these participants in the Seronegative Immunogenicity Analysis Set.

Table 10 Baseline characteristics (Seronegative Immunogenicity Analysis Set)

Characteristic/Statistic	V1222/ B1222 N = 342	V1222/ B2816 N = 341	VmRNA/ B1222 N = 294	VmRNA/ B2816 N = 294	Total N = 1271
Time since previous vaccination (days), n [a]					
n	342	341	294	294	1271
Mean	231.6	234.0	127.5	129.3	184.5
SD	92.3	89.0	28.2	28.9	86.6
Median	269.0	263.0	120.0	123.0	140.0
Min	74	91	71	73	71
Max	379	383	211	213	383
Primary vaccination dosing interval (days), n					
n	342	341	294	294	1271
Mean	54.2	52.4	63.4	62.3	57.7
SD	20.3	20.1	18.6	18.9	20.1
Median	59.0	56.0	70.0	70.0	64.0
Min	25	25	21	21	21
Max	91	98	86	89	98
BMI (kg/m^2), n					
n	341	341	293	294	1269
Mean	27.3	26.8	27.8	27.9	27.4
SD	5.2	4.7	5.8	6.1	5.4
Median	26.8	25.9	26.9	26.7	26.6
Comorbidity at baseline					
At least one [b]	158 (46.2)	156 (45.7)	138 (46.9)	143 (48.6)	595 (46.8)
BMI ≥ 30 kg/m^2	85 (24.9)	81 (23.8)	89 (30.3)	84 (28.6)	339 (26.7)
Significant CVD	84 (24.6)	92 (27.0)	61 (20.7)	61 (20.7)	298 (23.4)
Chronic lung disease	30 (8.8)	23 (6.7)	27 (9.2)	34 (11.6)	114 (9.0)
Diabetes	20 (5.8)	11 (3.2)	9 (3.1)	16 (5.4)	56 (4.4)
None	184 (53.8)	185 (54.3)	156 (53.1)	151 (51.4)	676 (53.2)

[a] Time since completion of the primary vaccination course (ie, second dose).
[b] At least one comorbidity vs no comorbidity, where comorbidity is BMI ≥ 30 kg/m^2 at baseline, Significant CVD, chronic lung disease, or diabetes
Percentages are based on N, the number of participants in the analysis set for each treatment group.
Comorbidity categories are derived from reported medical history and BMI.

A majority (>97%) of the participants in the AZD1222 booster treatment groups were enrolled in the UK. There are observed differences in demographics between the cohorts as a result of vaccination rollout timeline in the UK (June 2021 to October 2021). Starting in December 2020, the first phase of vaccination rollout in the UK prioritised the most vulnerable in a schedule primarily based on age and also prioritised healthcare workers, while all adults 18 years of age and older were able to get their first dose of a vaccine in June 2021. BNT162b2 was first deployed on 08 December 2020 and was administered at a 3-week interval; AZD1222 was first deployed on 04 January 2021. Participants from the mRNA cohort had received their primary vaccination series as part of the post-approval public rollout and would have been priority vaccine recipients (e.g. healthcare and social care workers). This explains the younger and more female participants in the mRNA cohort. Also, as the D7220C00001 study employed many of the same study sites as were used for the UK-based COV001 and COV002 studies of AZD1222, a significant number of participants from the AZD1222 cohort had also enrolled in these pre-approval studies. This may have resulted in longer interval between primary series vaccination and booster dose in the AZD1222 cohort. The Moderna mRNA-1273 vaccine was deployed in April 2021 in the UK, hence Study D7220C00001 includes only one seropositive participant that had received mRNA-1273 as primary series vaccination.

For matching criteria summary data for the historical control and AZD1222 cohorts, see Table 11.

Table 11 Matching criteria for historical controls and previously vaccinated AZD1222

	Statistic	HV1222(4) N = 508	V1222 N = 683
Age (years)	n	508	683
	Mean	59.4	60.1
	SD	13.65	13.44
	Median	62.0	62.0
Age group (years)			
18-64	n (%)	273 (53.7)	369 (54.0)
65 and older	n (%)	235 (46.3)	314 (46.0)
Sex			
Male	n (%)	266 (52.4)	370 (54.2)
Female	n (%)	242 (47.6)	313 (45.8)
BMI (kg/m2)	n	508	682
	Mean	27.0	27.0
	SD	4.83	4.95
	Median	26.3	26.3
Comorbidity at baseline			
At least one	n (%)	237 (46.7)	314 (46.0)
None	n (%)	271 (53.3)	369 (54.0)

Percentages are based on N, the number of subjects in the analysis set for each treatment group.
HV1222(4) is a historical control with a primary series of AZD1222 with a planned 4 week dosing interval.
The population of historical controls is selected from those who gave informed consent, received both vaccinations, had no protocol deviations judged to have the potential to interfere with the generation or interpretation of an immune response, had baseline and Day 29 post dose 2 pseudo neutralization data and prior to Day 29 post dose 2 had no prohibited concomitant medications, EUA vaccinations or positive PCR. These are compared to current participants in the seronegative immunogenicity analysis set.
Age groups are derived from the age groups specified at randomization. Sex is reported at randomization for historical controls and derived from the corrected value after randomization for current study. Comorbidity categories are derived from reported medical history and BMI.
BMI Body mass index; m2 Square meter; n Number of subjects in analysis for a continuous variable and number of subjects per category for a categorical variable; SD Standard deviation.
V1222 Previously vaccinated with AZD1222 and boosted with AZD1222 or AZD2816.

Immunogenicity Results for AZD1222 Booster Dose in Previously Vaccinated Cohort with Primary Series of AZD1222 or mRNA vaccine

The following sections summarises the humoral immunogenicity results in AZD1222 booster treatment groups of the previously vaccinated cohorts; AZD1222 cohort and mRNA cohort, describing the humoral immunogenicity of AZD1222 as a homologous and heterologous booster, respectively.

Baseline titres

Baseline neutralising antibody titres (both pseudoneutralising and S-protein binding antibodies) measured before administration of the booster dose varied widely within each treatment group. Overall, baseline neutralising antibodies were higher in participants previously vaccinated with an mRNA vaccine than participants previously vaccinated with AZD1222. In the AZD1222 cohort, baseline neutralising antibody titres were below the LLoQ in approximately 75% of participants as against the Wuhan-Hu-1 strain and approximately 87% as against the Beta variant. Corresponding values in the mRNA cohort were approximately 14% and 30%, respectively.

Table 12 Summary of participants with baseline pseudoneutralizing antibodies below the LLoQ (Seronegative Immunogenicity Set)

Strain	Statistics	V1222:B1222 (N=342)	V1222:B2816 (N=341)	VmRNA:B1222 (N=294)	VmRNA:B2816 (N=294)
B.1.351 (Beta)	n (%)	298 87.1%	297 87.1%	89 30.3%	83 28.2%
Wuhan-Hu-1	n (%)	257 75.1%	252 73.9%	41 13.9%	41 13.9%

AZD1222 Cohort (AZD1222 as a Homologous Booster)

Pseudoneutralising Antibodies Against Wuhan-Hu-1 and Beta in Seronegative Participants

Table 13 presents summary of descriptive GMT, GMFRs and seroresponse rate for pseudoneutralising antibody titres against Wuhan-Hu-1 strain and Beta variant in AZD1222 cohort that received a booster dose of AZD1222.

Table 13 Summary of Model-Adjusted GMT, GMFR, and seroresponse of pseudoneutralising antibodies in AZD1222 booster treatment group of previously vaccinated AZD1222 cohort (Seronegative Immunogenicity Analysis Set)

Statistic	V1222/B1222 N = 342				HV1222 N = 508	
	Wuhan-Hu-1 Strain		Beta Variant		Wuhan-Hu-1	
	Baseline	Day 29	Baseline	Day 29	Baseline	Day 29[a]
n	342	327	342	327	508	508
GMT (95% CI)	38.23 (34.82, 41.97)	248.89 (229.53, 269.89)	24.08 (22.00, 26.35)	184.96 (168.25, 203.33)	20.01 (19.00, 21.08)	242.80 (224.82, 262.23)
GMFR (95% CI)	--	6.54 (5.60, 7.63)	--	7.63 (6.43, 9.05)		12.13 (10.81, 13.61)
Seroresponse Rate (%) n (a) /N (b) (95% CI)	--	(66.1) 216 / 327 (60.6, 71.2)	--	(66.1) 216 / 327 (60.6, 71.2)		(84.1) 427/508 (80.6, 87.1)

[a] 28 days after second dose (or Day 57) of primary series in historical controls

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect. Age groups are derived from the age groups specified at randomization. Sex is derived from the corrected value after randomization. Comorbidity status is derived from the participant's medical history reported at screening.

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.

GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.

Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.

Seroresponse is defined as a ≥ 4-fold rise from baseline. Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.

Seroresponse in Seronegative Participants

The pseudoneutralising antibody seroresponse (ie, ≥ 4-fold increase in titres from baseline) to a booster dose of AZD1222 or AZD2816 in previously AZD1222 vaccinated participants is shown in Table 14. Seroresponse against Wuhan and beta strain generated by a booster dose of AZD1222 resulted in seroresponse rates of 66% in both cases.

Table 14 Summary of Model adjusted Seroresponse (Pseudoneutralising antibodies) in previously vaccinated participants (Seronegative immunogenicity Analysis Set)

	Day 15		Day 29	
Treatment group (strain)	**% (n/N)**	**95% CI**	**% (n/N)**	**95% CI**
V1222 cohort				
B1222 (Wuhan-Hu-1)	61.4 (204 / 332)	56.0, 66.7	66.1 (216 / 327)	60.6, 71.2
B2816 (Wuhan-Hu-1)	61.7 (195 / 316)	56.1, 67.1	65.8 (210 / 319)	60.3, 71.0
B1222 (Beta)	64.2 (213 / 332)	58.7, 69.3	66.1 (216 / 327)	60.6, 71.2
B2816 (Beta)	80.1 (253 / 316)	75.2, 84.3	82.8 (264 / 319)	78.2, 86.7

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.

Seroresponse is defined as a ≥ 4-fold rise from baseline.

Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.

Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.

Non-Inferiority of Immune Response (GMT Ratio and Difference in Seroresponse Rate) in AZD1222 Booster Treatment Group of the AZD1222 Cohort (Homologous AZD1222 Booster)

The Wuhan-Hu-1 pseudoneutralising antibody GMT ratio at Day 29 after AZD1222 booster to that at Day 29 after dose 2 of primary series in historical control was 1.03 (95% CI 0.917, 1.146), which met the 1.5-fold non-inferiority criterion (ie, lower bound of the 2-sided 95% CI for GMTR >0.67).

The prespecified non-inferiority criterion for difference in seroresponse rate against Wuhan-Hu-1 was not met (lower bound of 95% CI = −24.0%) (Table 15).

Table 15 Non-inferiority analysis of GMT Ratio and Seroresponse rate at Day 29 after AZD1222 booster in AZD1222 treatment group of previously vaccinated AZD1222 cohort (Seronegative Immunogenicity Analysis Set)

Endpoint: Comparator vs. Reference	**Statistic**	**GMT Ratio**	**Difference in Seroresponse**
Primary[a]**:** V1222/ B1222 (Wuhan) vs. HV1222 (Wuhan)	Ncomp/Nref	327/508	
	Value	1.03	-18.0
	95% CI[b]	(0.917, 1.146)	(-24.0, -12.0)
Other Secondary: V1222/B1222 (Beta) vs. HV1222 (Wuhan)	Ncomp/Nref	327/508	
	Value	0.76	-18.0
	95% CI[b]	(0.674, 0.861)	(-24.0, -12.0)
Other Secondary: V1222/B1222 (Beta) vs. V1222/B1222 (Wuhan)	Ncomp/Nref	327/327	
	Value	0.74	0.0
	95% CI[c]	(0.656, 0.842)	(-7.2, 7.2)

[a] Non-inferiority of GMT ratio is the primary endpoint. Difference in seroresponse is a other secondary endpoint

[b] The CI assumes unequal variances as the equality of variance assumption was rejected at alpha = 0.05

[c] The CI assumes equal variances as the equality of variance assumption was not rejected

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ.

Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.

The fold ratio is calculated as the ratio of the titre levels in the comparator arm to the reference arm. The difference in seroresponse is calculated as (seroresponse rate of comparator arm) - (seroresponse rate of reference arm), where the seroresponse rate is calculated from those who have titre assessments at baseline and the time point of interest. The CI for the difference in seroresponse is calculated using the Newcombe method based on the Wilson score.

Seroresponse is defined as a ≥ 4-fold rise from baseline.

B1222: Participants receiving a third dose booster of AZD1222; CI: Confidence interval; HV1222: Historical controls vaccinated with a primary series of AZD1222; Ncomp: Number of subjects with assessments at given time point in the comparator group; Nref: Number of subjects with assessments at given time point in the reference group; GMTR: Geometric mean titre ratio; LLoQ: Lower limit of quantitation; SRR: Seroresponse rate; ULoQ: Upper limit of quantitation; V1222: Participants previously vaccinated with a primary series of AZD1222.

Although the GMT of pseudoneutralising antibodies against Wuhan-Hu-1 at Day 29 after boosting was similar to that at Day 29 after second dose of primary series in historical control (248.89 vs 242.80), the baseline GMT against Wuhan-Hu-1 strain were approximately twice as high in participants from AZD1222 cohort as compared to the historical controls (38.23 vs 20.01, respectively). It is not surprising that the proportion of participants exhibiting the greater than 4-fold increase was lower following a booster of AZD1222 compared with a primary vaccination in the historical control (66.1 % and 84.1 %) (Table 13), however the GMT results support the clinical significance of the increase seen with the booster dosing.

In previously vaccinated participants in the AZD1222 booster treatment group who were seropositive at baseline in Study D7220C00001, the baseline neutralising antibody GMTs against Wuhan-Hu-1 were higher compared to those in seronegative participants. At Day 29, following AZD1222 booster, lower GMFRs and seroresponse rate were noted in seropositive participants, however the absolute values of GMTs against Wuhan-Hu-1 and Beta were notably higher. For example, the GMFR for pseudoneutralising antibodies against Wuhan-Hu-1 was 2.59 and seroresponse rate was 36.8% in seropositive participants as compared to 6.54 and 66.1% in seronegative participants, respectively.

Spike-Binding Antibodies Against Wuhan-Hu-1 Strain and Beta, Alpha, and Gamma Variants

At Day 29 after an AZD1222 booster, a robust and broad humoral response was elicited as measured by spike-binding antibody titres using a multiplex ECL assay against Wuhan-Hu-1 strain and Beta, Alpha and Gamma variants, representing a 9.43, 10.29, 10.09, and 10.05 fold-rise from baseline, respectively.

The S-protein binding antibody seroresponse (ie, ≥ 4-fold increase in titre from baseline) to a booster dose of AZD1222 or AZD2816 is shown in Table 16. In Table 17 it is shown that the seroresponse rate of the historic control group, based on S-binding antibodies, was 98.8% at day 29 after second doses of AZD1222.

Table 16 Summary of Model adjusted Seroresponse (S-Protein binding antibodies) in previously vaccinated participants (seronegative immunogenicity analysis set)

	Day 15		Day 29	
Treatment group (strain)	**% (n/N)**	**95% CI**	**% (n/N)**	**95% CI**
V1222				
B1222 (Wuhan-Hu-1)	65.5 (218 / 333)	60.1, 70.6	68.2 (219 / 321)	62.8, 73.3
B2816 (Wuhan-Hu-1)	62.4 (199 / 319)	56.8, 67.7	63.8 (201 / 315)	58.2, 69.1
B1222 (Beta)	69.1 (230 / 333)	63.8, 74.0	71.0 (228 / 321)	65.7, 75.9
B2816 (Beta)	74.3 (237 / 319)	69.1, 79.0	75.2 (237 / 315)	70.1, 79.9

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.
Seroresponse is defined as a ≥ 4-fold rise from baseline.
Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
B1222 or B2816, Participants receiving a third dose booster of AZD1222 or AZD2816; CI, Confidence interval; GMT, Geometric mean titre; n, Number of subjects in analysis; N, Number of subjects per treatment group; V1222, Participants previously vaccinated with 2 doses of AZD1222; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine
Source: Table 14.2.2.1

Table 17 Summary of Model adjusted GMT, GMFR and seroresponse for historical cohort

Cohort: HV1222(4)
Treatment: AZD1222
Assay: Spike protein binding
Strain: Wuhan-Hu-1

		Day 29 Post Dose 1		Day 29 Post Dose 2	
Statistic	**Baseline**	**Observed**	**Fold Rise**	**Observed**	**Fold Rise**
n	508	352	352	508	508
GMT	49.63	5893.06	110.87	19639.22	395.72
95% CI	(45.40, 54.25)	(5414.50, 6413.92)	(94.10, 130.62)	(18208.38, 21182.49)	(344.53, 454.52)
Min	5.7	246.0	0.8	29.0	0.1
Max	1483.6	74961.7	7992.4	649413.9	61557.8
log(GMT)	5.63	12.52	6.79	14.26	8.63
SD	1.474	1.166	2.256	1.252	2.293
95% CI	(5.50, 5.76)	(12.40, 12.65)	(6.56, 7.03)	(14.15, 14.37)	(8.43, 8.83)
Seroresponse n(a)/N(b) (%)			337 / 352 (95.7)		502 / 508 (98.8)
95% CI			(93.1, 97.6)		(97.4, 99.6)

a. n is the number of seroresponders.
b. N is the number of subjects with a baseline measurement and an assessment at the given time point.
HV1222(4) is a historical control with a primary series of AZD1222 with a planned 4 week dosing interval.
The population of historical controls is selected from those who gave informed consent, received both vaccinations, had no protocol deviations judged to have the potential to interfere with the generation or interpretation of an immune response, had baseline and Day 29 post dose 2 pseudo neutralization data and prior to Day 29 post dose 2 had no prohibited concomitant medications, COV vaccinations or positive PCR.
Seroresponse is defined as a >= 4-fold rise from baseline.
Estimation performed using a linear model with the log transformed value of the titer as the dependent variable, independent variables for visit window, baseline comorbidities (At least one or None), sex (Male or Female) and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.
Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titer levels to calculate seroresponse.
Titer values measured as below LLoQ (33) are imputed to a value that is half of the LLoQ. Titer values measured as above ULoQ (2,000,000) are imputed at the ULoQ value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titer.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
CI confidence interval; GMFR geometric mean fold rise; GMT geometric mean titer; n Number of subjects in analysis;
SD Standard deviation; NE Not Evaluable; LLoQ Lower limit of quantitation; ULoQ Upper limit of quantitation.

mRNA Cohort (AZD1222 as a Heterologous Booster)

Pseudoneutralising Antibodies Against Wuhan-Hu-1 Strain and Beta Variant

Table 18 presents GMT, GMFR and seroresponse rate for pseudoneutralising antibodies against Wuhan-Hu-1 strain and Beta variant in the previously mRNA vaccinated cohort.

Table 18 Summary of Model-Adjusted GMT, GMFR and Seroresponse of Pseudoneutralising antibodies in AZD1222 booster treatment group of previously vaccinated mRNA cohort (Seronegative immunogenicity Analysis Set)

Statistic	VmRNA/B1222				HV1222	
	Wuhan-Hu-1 Strain		Beta Variant		Wuhan-Hu-1	
	Baseline	Day 29	Baseline	Day 29	Baseline	Day 29[a]
n	294	278	294	278	508	508
GMT (95% CI)	197.74 (179.59, 217.72)	747.18 (699.46, 798.16)	106.81 (96.81, 117.84)	637.42 (593.75, 684.30)	20.01 (19.00, 21.08)	242.8 (224.82, 262.23)
GMFR (95% CI)	--	3.77 (3.26, 4.37)	--	5.93 (5.07, 6.93)	--	12.13 (10.81, 13.61)
SRR (%) n (a) /N (b) (95% CI)	--	(43.2) 120 / 278 (37.3, 49.2)	--	(57.6) 160 / 278 (51.5, 63.4)	--	(84.1) 427/508 (80.6, 87.1)

[a] 28 days after second dose (or Day 57) of primary series in historical controls

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.

GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.

Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.

Seroresponse is defined as a ≥ 4-fold rise from baseline. Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.

Seroresponse in Seronegative Participants

The pseudo neutralising antibody seroresponse (i.e., ≥ 4-fold increase in titres from baseline) to a booster dose of AZD1222 or AZD2816 in previously mRNA vaccinated participants is shown in Table 19. Seroresponse against Wuhan strain and Beta variant generated by a booster dose of AZD1222 resulted in seroresponse rates of 43.2% to 57.6%, respectively.

Table 19 Summary of Model adjusted seroresponse (Pseudoneutralising Antibodies) in previously vaccinated participants (Seronegative immunogenicity Analysis Set)

	Day 15		Day 29	
Treatment group (strain)	**% (n/N)**	**95% CI**	**% (n/N)**	**95% CI**
VmRNA cohort				
B1222 (Wuhan-Hu-1)	35.4 (97 / 274)	29.7, 41.4	43.2 (120 / 278)	37.3, 49.2
B2816 (Wuhan-Hu-1)	41.1 (113 / 275)	35.2, 47.2	49.6 (137 / 276)	43.6, 55.7
B1222 (Beta)	54.4 (149 / 274)	48.3, 60.4	57.6 (160 / 278)	51.5, 63.4
B2816 (Beta)	77.5 (213 / 275)	72.1, 82.3	80.4 (222 / 276)	75.3, 84.9

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.

Seroresponse is defined as a ≥ 4-fold rise from baseline.

Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.

Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.

Non-Inferiority of Immune Response (GMT Ratio and Difference in Seroresponse Rate) in AZD1222 Booster Treatment Group of mRNA cohort (Heterologous AZD1222 Booster)

Immunobridging of spike-binding and neutralising antibody data in participants heterologously boosted with AZD1222 after a primary series of an mRNA vaccine to those vaccinated with a primary series of AZD1222 allows the comparison of neutralising antibody titres post-boost to a regimen that is shown to be clinically protective after a primary series. Hence the matched historical controls from study D8110C00001 were appropriately used as a reference arm in analysis of data from the mRNA cohort.

The Wuhan-Hu-1 pseudoneutralising antibody GMT ratio at Day 29 after AZD1222 booster in the mRNA cohort to that at Day 29 after dose 2 of primary series in historical control was 3.08 (95% CI 2.781, 3.405), which met the 1.5-fold non-inferiority criterion (ie, lower bound of the 2-sided 95% CI for GMTR >0.67) (Table 20)

The prespecified non-inferiority criterion for difference in seroresponse rate against Wuhan-Hu-1 was not met (lower bound of 95% CI = -47.3%). This is attributed to the higher baseline GMT values in previously vaccinated participants as compared the matched unvaccinated controls (197.74 versus 20.01) (Table 18).

Table 20 Non-inferiority analysis of GMT Ratio and Seroresponse Rate at Day 29 after AZD1222 booster in AZD1222 treatment group of previously vaccinated mRNA cohort (Seronegative immunogenicity analysis Set)

Endpoint: Comparator vs. Reference	Statistic	GMT Ratio	Difference in Seroresponse
Primary[a]: VmRNA/ B1222 (Wuhan) vs. HV1222 (Wuhan)	Ncomp/Nref	278/508	
	Value	3.08	-40.9
	95% CI[b]	(2.781, 3.405)	(-47.3, -34.1)
Other Secondary: VmRNA /B1222 (Beta) vs. HV1222 (Wuhan)	Ncomp/Nref	278/508	
	Value	2.63	-26.5
	95% CI[b]	(2.365, 2.914)	(-33.1, -19.9)
Other Secondary: VmRNA/B1222 (Beta) vs. VmRNA/B1222 (Wuhan)	Ncomp/Nref	278/278	
	Value	0.85	14.4
	95% CI[c]	(0.774, 0.940)	(6.1, 22.4)

[a] Non-inferiority of GMT ratio is the primary endpoint. Difference in seroresponse is a secondary endpoint

[b] The CI assumes unequal variances as the equality of variance assumption was rejected at alpha = 0.05

[c] The CI assumes equal variances as the equality of variance assumption was not rejected.

Seroresponse is defined as a ≥ 4-fold rise from baseline.

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ.

Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.

The fold ratio is calculated as the ratio of the titre levels in the comparator arm to the reference arm. The difference in seroresponse is calculated as (seroresponse rate of comparator arm) - (seroresponse rate of reference arm), where the seroresponse rate is calculated from those who have titre assessments at baseline and the time point of interest. The CI for the difference in seroresponse is calculated using the Newcombe method based on the Wilson score.

Spike-binding Antibodies Against Wuhan-Hu-1 Strain and Beta Variant

At Day 29 after a AZD1222 booster in the previously mRNA vaccinated cohort, a broad humoral response was elicited as assessed by quantifying the spike-binding antibodies by a multiplex ECL assay against Wuhan-Hu-1, Beta, Alpha and Gamma variants with a 3.24, 3.02, 3.37, and 3.08 fold-rise from baseline, respectively. The proportion of participants with seroresponse to spike-binding antibodies to these variants were also comparable.

Overall, the GMFR and seroresponse rates of spike-binding antibodies to Wuhan-Hu-1 strain and Beta variant at Day 29 after a AZD1222 booster were lower in the mRNA cohort as compared to the AZD1222 cohort.

A robust S-protein binding antibody seroresponse (ie, ≥ 4-fold increase in titre from baseline) to a booster dose of AZD1222 or AZD2816 was observed in the AZD1222 cohort, with reduced seroresponse observed in the mRNA cohort. Table 19 presents data for both cohorts.

Table 19 **Summary of Model Adjusted Seroresponse (S-Protein Binding Antibodies) in Previously Vaccinated Participants (Seronegative Immunogenicity Analysis Set)**

	Day 15		Day 29	
Treatment group (strain)	% (n/N)	95% CI	% (n/N)	95% CI
V1222				
B1222 (Wuhan-Hu-1)	65.5 (218 / 333)	60.1, 70.6	68.2 (219 / 321)	62.8, 73.3
B2816 (Wuhan-Hu-1)	62.4 (199 / 319)	56.8, 67.7	63.8 (201 / 315)	58.2, 69.1
B1222 (Beta)	69.1 (230 / 333)	63.8, 74.0	71.0 (228 / 321)	65.7, 75.9
B2816 (Beta)	74.3 (237 / 319)	69.1, 79.0	75.2 (237 / 315)	70.1, 79.9
VmRNA				
B1222 (Wuhan-Hu-1)	29.9 (82 / 274)	24.6, 35.7	36.7 (99 / 270)	30.9, 42.7
B2816 (Wuhan-Hu-1)	35.0 (96 / 274)	29.4, 41.0	40.0 (108 / 270)	34.1, 46.1
B1222 (Beta)	27.0 (74 / 274)	21.8, 32.7	31.1 (84 / 270)	25.6, 37.0
B2816 (Beta)	42.6 (116 / 272)	36.7, 48.8	49.6 (133 / 268)	43.5, 55.8

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.
Seroresponse is defined as a ≥ 4-fold rise from baseline.
Model-adjusted estimates for seroresponse are derived using the model-adjusted baseline and post-baseline titre levels to calculate seroresponse.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
B1222 or B2816, Participants receiving a third dose booster of AZD1222 or AZD2816; CI, Confidence interval; GMT, Geometric mean titre; n, Number of subjects in analysis; N, Number of subjects per treatment group; V1222, Participants previously vaccinated with 2 doses of AZD1222; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine
Source: Table 14.2.2.1

Other secondary and exploratory endpoints

Sensitivity analysis

Table 21 is a summary of the primary analysis results for each of the sensitivity analyses.

Table 21 GMT and seroresponse comparisons of the primary endpoints for the primary analysis and the sensistivity analyses (Seronegative immunogenicity analysis set)

Table 1 **GMT and Seroresponse Comparisons of the Primary Endpoints for the Primary Analysis and the Sensitivity Analyses (Seronegative Immunogenicity Analysis Set)**

Comparator vs. Reference	Statistic	Primary Analysis		Adjusting for Continuous Age		Adjusting for Continuous Age and BMI		Adjusting for Continuous Dose Interval	
		GMT Ratio	Seroresponse difference	GMT Ratio	Seroresponse difference	GMT Ratio	Seroresponse difference	GMT Ratio	Seroresponse difference
Primary: V1222/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	327/508		327/508		326/508		327/508	
	Value	1.03	-18.0	1.01	-18.0	1.01	-18.1	1.04	-18.0
	95% CI[a]	0.92, 1.15	-24.0, -12.0	0.90, 1.13	-24.0, -12.0	0.91, 1.13	-24.2, -12.1	0.93, 1.16	-24.0, -12.0
Primary: VmRNA/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	278/508		278/508		277/508		278/508	
	Value	3.08	-40.9	3.10	-40.9	3.09	-41.1	2.90	-40.9
	95% CI[a]	2.78, 3.41	-47.3, -34.1	2.80, 3.43	-47.3, -34.1	2.80, 3.42	-47.5, -34.3	2.61, 3.20	-47.3, -34.1

[a] Confidence intervals for the comparison assumes unequal variances as the equality of variance assumption was rejected at alpha = 0.05.
Seroresponse is defined as a ≥ 4-fold rise from baseline.
The population of historical controls is selected from those who gave informed consent, received both vaccinations, had no protocol deviations judged to have the potential to interfere with the generation or interpretation of an immune response, had baseline and Day 57 pseudoneutralisation data and prior to Day 57 had no prohibited concomitant medications, EUA vaccinations or positive PCR. These are compared to current study participants in the seronegative immunogenicity analysis set.
The fold ratio is calculated as the ratio of the titre levels in the comparator arm to the reference arm. The difference in seroresponse is calculated as (seroresponse rate of comparator arm) - (seroresponse rate of reference arm), where the seroresponse rate is calculated from those who have titre assessments at baseline and the time point of interest. The CI for the difference in seroresponse is calculated using the Newcombe method based on the Wilson score.
B1222, Participants receiving a third dose booster of AZD1222; CI, Confidence interval; GMT, Geometric mean titres; n, Number of subjects in analysis; V1222, Participants previously vaccinated with 2 doses of AZD1222; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine.
Derived from: IEMT Tables 6.1.3.1, 6.2.3.1, and 6.3.3.1, and Interim CSR Tables 14 and 15

Exploratory Immunogenicity Assessment Against Delta Variant in the AZD1222 Booster Treatment Group of the Previously Vaccinated Cohorts (AZD1222 Cohort and mRNA Cohort)

In an exploratory analysis, the humoral response against the Delta variant was conducted. An alternative

platform of pseudovirus neutralisation assay was utilised to assess neutralising responses to this variant. Overall, homologous and heterologous AZD1222 booster resulted in comparable fold rise in the GMT of pseudoneutralising antibodies over baseline against the Delta variant. These exploratory data are limited due the use of an unvalidated pseudoneutralisation assay and analysis was conducted on a small subset of participants.

Table 22 presents GMT, GMFR and seroresponse rate for pseudoneutralising antibodies against Delta variant in previously vaccinated cohorts.

Table 22 Summary of Model-Adjusted GMT, GMFR and seroresponse of Pseudoneutralising antibodies against delta variant in AZD1222 booster treatment group of previously vaccinated cohorts (Seronegative Immunogenicity Analysis Set)

Statistic	V1222/B1222 N = 342		VmRNA/B1222 N = 294	
	Baseline	Day 29	Baseline	Day 29
n	99	91	95	81
GMT (95% CI)	21.82 (19.45, 24.48)	61.24 (52.78, 71.07)	53.85 (46.99, 61.73)	163.43 (141.60, 188.62)
n		91		80
GMFR (95% CI)	--	2.79 (2.27, 3.42)	--	3.12 (2.48, 3.92)

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.

Titre values measured as below LLoQ (50) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (12,150) are imputed at the ULoQ value.

GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.

Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.

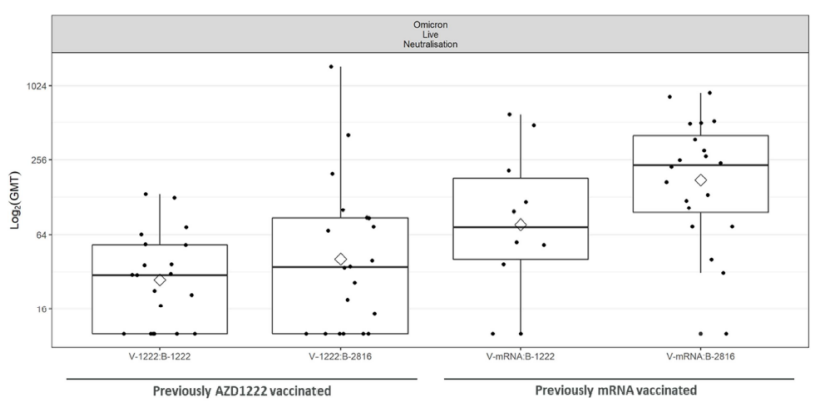
Exploratory Immunogenicity Assessment Against Omicron Variant in the AZD1222 Booster Treatment Group of the Previously Vaccinated Cohorts (AZD1222 Cohort and mRNA Cohort)

In a separate CSR (study MS1222-0007), the anti-omicron antibodies present in subjects boosted with AZD1222, who were previously vaccinated with either AZD1222 or an mRNA vaccine previously, was analysed. The different assays were performed: the University of Oxford, Omicron live virus neutralisation (Oxford, UK) and the UKHSA Omicron Live virus neutralisation (Porton Down, UK).

Results of Omicron live virus neutralisation assay, University of Oxford

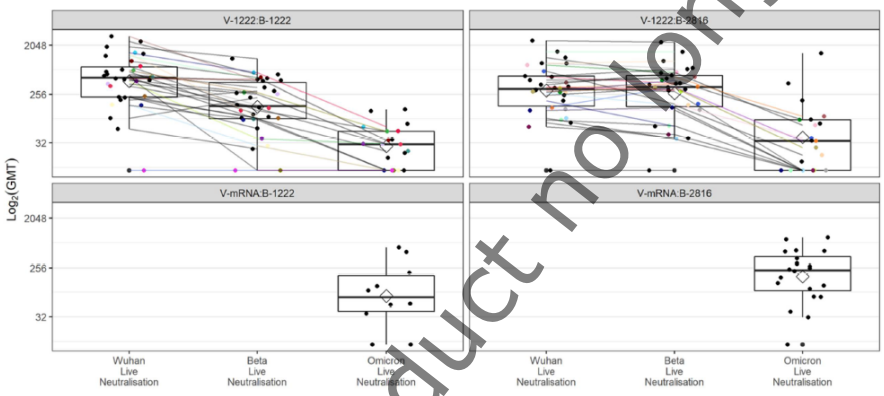
Neutralising antibody responses from serum samples from the D7220C00001 study were assessed 28 days after a booster dose of AZD1222 or AZD2816 in participants from previously vaccinated with 2 doses of AZD1222 or an mRNA vaccine (Figure 5). Neutralising antibody responses against the Omicron variant were detected following a boost with AZD1222 in the majority of study participants, with numerically higher GMTs detected in the heterologous boost group. As expected, nAb titres against the Omicron variant were lower than those observed against either the ancestral Wuhan-Hu-1 strain or Beta variant for participants previously vaccinated with AZD1222 (Figure 6).

Figure 5 Neutralising antibody responses to the Omicron variant 28 days post boost



B1222: received a booster dose of AZD1222; B2816: received a booster dose of AZD2816; GMT: geometric mean titres; V1222: vaccinated with 2 doses of AZD1222; VmRNA: vaccinated with 2 doses of an mRNA COVID-19 vaccine

Figure 6 Neutralising antibody responses to the Wuhan-hu-1 strain, Beta and Omicron variants 28 days post boost



B1222: received a booster dose of AZD1222; B2816: received a booster dose of AZD2816; GMT: geometric mean titres; V1222: vaccinated with 2 doses of AZD1222; VmRNA: vaccinated with 2 doses of an mRNA COVID-19 vaccine.

Results of Omicron live virus neutralisation assay, UKHSA.

Table 23 shows an analysis conducted by the UKHSA that presents similar data to Figure 5 but include pre-booster dose assessments against the Omicron variant. In the study D7220C00001 cohorts from which sera were analysed, the median interval between boost and primary series was approximately 9 months for the group previously vaccinated with AZD1222 and approximately 4 months for the group previously vaccinated with mRNA.

Given the long interval since primary series, all analysed study participants in the group previously vaccinated with AZD1222 had baseline titres below the lower limit of quantification. AZD1222 induced increases in nAb titres to the Omicron variant over baseline.

Table 23 Summary statistics of Omicron live virus neutralisation assay performed at the UKHSA

Visit	Statistic	V1222:B1222	V1222:B2816	VmRNA:B1222	VmRNA:B2816
Baseline	**n**	**10**	**10**	**9**	**8**
	GMT	10.0	10.0	25.6	21.5
	95% CI for GMT	NE, NE	NE, NE	14.68, 44.65	12.55, 36.90
	Min, Max	10, 10	10, 10	10, 52	10, 40
	Median	10	10	40	29
Day 29	**n**	**10**	**10**	**10**	**10**
	GMT	36.1	74.7	85.1	161.2
	95% CI for GMT	17.70, 73.69	32.15, 173.75	44.99, 161.10	89.33, 290.75
	Min, Max	10, 115	10, 636	40, 471	40, 479
	Median	49	61	60	177

B1222: received a booster dose of AZD1222; B2816: received a booster dose of AZD2816; CI=confidence interval; GMT: geometric mean titres; NE: not evaluated; Min: minimum; Max: maximum; V1222: vaccinated with 2 doses of AZD1222; VmRNA: vaccinated with 2 doses of an mRNA COVID-19 vaccine

Subgroup Results

Increases in humoral immunogenicity were observed for all prespecified subgroups of age, sex, and comorbidity following booster doses of AZD2816 or AZD1222 in both the V1222 and VmRNA seronegative cohorts. Overall, the responses were similar across subgroups. The following numerical differences were noted:

- Pseudoneutralising responses (fold rise and seroresponse) were numerically lower in adults ≥ 65 years of age than in younger adults in the AZD1222 cohort but the opposite was observed for the mRNA cohort. S-protein binding antibody responses were similar across age groups within both the AZD1222 and mRNA cohorts.
- Humoral immunogenicity was similar between male and female participants, with only modest numerically increased pseudoneutralising and binding responses for most variants in female participants.
- Humoral immunogenicity, including to variants of concern, was not decreased in participants with at least one comorbidity as assessed by S-protein binding antibodies and pseudoneutralising antibodies.

Cell-Mediated Immunogenicity

T cell responses were assessed in an IFNγ ELISpot assay with peptides specific to the Wuhan- Hu-1 strain (Wuhan S1 + S2). Modest increases in Spike-specific IFNγ T Cell responses were observed following a booster dose of AZD1222 or AZD2816 at Day 15 post-booster (Table 24). A limitation to this analysis was the small subgroup of participants.

Table 24 Summary of Model Adjusted GMR and GMFR for ELISpot responses (Seronegative Immunogenicity Analysis Set)

	Baseline	Day 15		Day 29	
Statistic		**GMR**	**Fold rise**	**GMR**	**Fold rise**
V1222/B1222 (Wuhan S1 + S2 assay) N=342					
n	19	23	18	22	18
GMR	20.54	34.89	1.69	21.50	1.06
95% CI	16.36, 25.80	28.74, 42.36	1.15, 2.48	17.52, 26.39	0.68, 1.64
V1222/B2816 (Wuhan S1 + S2 assay) N=341					
n	9	11	8	12	8
GMR	28.08	43.73	1.82	33.99	1.09
95% CI	15.07, 52.32	27.53, 69.48	0.54, 6.16	23.55, 49.06	0.38, 3.15
VmRNA/B1222 (Wuhan S1 + S2 assay) N=294					
n	23	24	20	22	18
GMR	88.36	151.95	1.79	97.54	1.10
95% CI	75.79, 103.01	126.77, 182.14	1.28, 2.50	81.02, 117.43	0.78, 1.56
VmRNA/B2816 (Wuhan S1 + S2 assay) N=294					
n	19	21	18	20	17
GMR	82.73	105.16	1.24	108.44	1.34
95% CI	63.47, 107.83	81.84, 135.12	0.77, 1.99	82.83, 141.96	0.81, 2.23

Estimation performed using a linear model with the log transformed value of the titre as the dependent variable, independent variables for visit window (Baseline, Day 15, Day 29), time since previous vaccination, baseline comorbidities (At least one or None), sex (Male or Female), and age group (18-64 or 65 and older) as fixed effects, and participant as a random effect.

Values measured as below LLoQ (16) are imputed to a value that is half of the LLoQ.

GMR is calculated as the antilogarithm transformation of the mean of the log-transformed result.

B1222 or B2816, Participants receiving a third dose booster of AZD1222 or AZD2816; CI, Confidence interval; ELISpot, Enzyme-linked immune absorbent spot; GMFR, Geometric mean fold rise; GMR, Geometric mean response; LLoQ, Lower limit of quantification; n, Number of subjects in analysis; N, Number of subjects per treatment group; V1222, Participants previously vaccinated with 2 doses of AZD1222; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine

Immunogenicity (Anti-Vector)

Anti-vector responses following a booster dose of AZD1222 and AZD2816 were evaluated by a validated bioanalytical method in seronegative study participants. In the AZD1222 cohort, most participants had pre-existing anti-vector responses at baseline, which increased following a third dose booster. See Table 25.

Table 25 Summary of GMT, GMFR and seroresponse by age group (Seronegative immunogenicity analysis set)

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Table 14.2.1.3 Summary of GMT, GMFR and seroresponse by Age Group (Seronegative immunogenicity analysis set)

Cohort: V1222
Treatment: B1222
Assay: ChAdOx1 nAB
Strain: N/A
Age group: 18-64

	Baseline	Day 15		Day 29	
Statistic	(N=185)	Observed	Fold Rise	Observed	Fold Rise
n	172	158	156	141	139
GMT	593.24	4060.78	6.86	4452.70	7.47
95% CI	(483.75, 727.51)	(3227.54, 5109.12)	(5.33, 8.82)	(3521.20, 5630.62)	(5.80, 9.61)
Min	20.0	20.0	0.0	138.0	0.4
Max	20029.0	146730.0	710.6	254608.0	376.9
log(GMT)	9.21	11.99	2.78	12.12	2.90
SD	1.956	2.108	2.294	2.034	2.170
95% CI	(8.92, 9.51)	(11.66, 12.32)	(2.42, 3.14)	(11.78, 12.46)	(2.54, 3.26)
Seroresponse n(a)/N(b) (%)			90 / 156 (57.7)		83 / 139 (59.7)
95% CI			(49.5, 65.6)		(51.1, 67.9)

a. n is the number of seroresponders.
b. N is the number of subjects with a baseline measurement and an assessment at the given time point.
Age groups are derived from the age groups specified at randomization.
Seroresponse is defined as a >= 4-fold rise from baseline.
Titer values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titer values measured as above ULoQ (787,339) are imputed at the ULoQ value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titer.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
CI confidence interval; GMFR geometric mean fold rise; GMT geometric mean titer; n Number of subjects in analysis;
N Number of subjects per treatment group and age group; SD Standard deviation; NE Not Evaluable; LLoQ Lower limit of quantitation;
ULoQ Upper limit of quantitation; V primary vaccination; B booster vaccination.

Source data: ADIS eDC Extraction Date 2021-10-11
Program: /data/prod/client04/d7220c00001/ia2_unblinded/programs/statout/tl4020103.sas (2022-01-28 21:52:04) Final Page 15 of 64

In the mRNA cohort, where baseline anti-ChAdOx1 neutralising antibodies were low, GMTs at Day 29 after a single booster dose of AZD1222 or AZD2816 were similar to those observed at baseline in the previously AZD1222 vaccinated treatment groups. See Table 26.

Table 26 Summary of GMT, GMFR and seroresponse by age group (Seronegative immunogenicity analysis set)

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Table 14.2.1.3 Summary of GMT, GMFR and seroresponse by Age Group (Seronegative immunogenicity analysis set)

Cohort: VmRNA
Treatment: B1222
Assay: ChAdOx1 nAB
Strain: N/A
Age group: 18-64

	Baseline	Day 15		Day 29	
Statistic	(N=215)	Observed	Fold Rise	Observed	Fold Rise
n	202	183	181	183	181
GMT	25.36	1551.59	75.28	1154.22	46.63
95% CI	(23.15, 27.78)	(1412.49, 2586.57)	(56.86, 99.68)	(882.29, 1509.95)	(35.94, 60.50)
Min	20.0	20.0	1.0	20.0	1.0
Max	588.0	787339.0	39367.0	787339.0	39367.0
log(GMT)	4.66	10.93	6.23	10.17	5.54
SD	0.949	2.786	2.761	2.657	2.561
95% CI	(4.53, 4.80)	(10.52, 11.34)	(5.83, 6.64)	(9.79, 10.56)	(5.17, 5.92)
Seroresponse n(a)/N(b) (%)			172 / 181 (95.0)		169 / 181 (93.4)
95% CI			(90.8, 97.7)		(88.7, 96.5)

a. n is the number of seroresponders.
b. N is the number of subjects with a baseline measurement and an assessment at the given time point.
Age groups are derived from the age groups specified at randomization.
Seroresponse is defined as a >= 4-fold rise from baseline.
Titer values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titer values measured as above ULoQ (787,339) are imputed at the ULoQ value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titer.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
CI confidence interval; GMFR geometric mean fold rise; GMT geometric mean titer; n Number of subjects in analysis;
N Number of subjects per treatment group and age group; SD Standard deviation; NE Not Evaluable; LLoQ Lower limit of quantitation;
ULoQ Upper limit of quantitation; V primary vaccination; B booster vaccination.

Source data: ADIS eDC Extraction Date 2021-10-11
Program: /data/prod/client04/d7220c00001/ia2_unblinded/programs/statout/tl4020103.sas (2022-01-28 21:52:04) Final Page 47 of 64

Pairwise correlative analyses between anti-vector responses and pseudoneutralising antibody responses

to both the Wuhan-Hu-1 strain as well as the Beta variant were conducted in order to assess the impact of pre-existing anti-vector immunity on Spike specific immunogenicity. Overall, minimal correlation was observed between ChAdOx1 pseudoneutralising antibody titres and pseudoneutralising antibody responses to SARS-CoV-2 (Table 27).

Table 27 Pearson correlation: Day 29 Titres vs anti -vector baseline titres (Seronegative Immunogenicity Analysis Set)

	N	Pearson Correlation	p-value
Pseudo nAb - Wuhan (1/Dilution)			
V1222/B1222	307	-0.08415	0.1413
V1222/B2816	310	-0.02246	0.6936
VmRNA/B1222	263	-0.04234	0.4942
VmRNA/B2816	263	-0.05611	0.3648
Pseudo nAb - Beta (1/Dilution)			
V1222/B1222	307	-0.03885	0.4977
V1222/B2816	310	-0.00720	0.8995
VmRNA/B1222	263	-0.06759	0.2748
VmRNA/B2816	263	-0.04052	0.5130

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.

Correlations are based on log2 titre values.

N number of subjects with data for both assays at the applicable timepoints.

B1222 or B2816, Participants receiving a third dose booster of AZD1222 or AZD2816; LLoQ, Lower limit of quantification; nAb, Neutralising antibodies; N, Number of subjects per treatment group; ULoQ, Upper limit of quantification; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222.

Source: see IEMT Table 4 in Section 14.2

Results for Seropositive Participants

Table 28 presents pseudoneutralising antibody data for AZD1222-cohort participants who were seropositive at baseline. Raw values are presented; there were too few participants to perform a model-adjusted analysis.

Table 28 Summary of Pseudoneutralising antibody GMT and GMFR in participants previously vaccinated with AZD1222 (Seropositive immunogenicity Analysis set)

	Baseline	Day 15		Day 29	
Statistic		GMT	Fold rise	GMT	Fold rise
V1222/B1222 (Wuhan-Hu-1) N=20					
n	20	18	18	19	19
GMT	138.51	405.13	2.73	346.27	2.59
95% CI	49.53, 387.34	168.79, 972.40	1.59, 4.68	155.96, 768.79	1.42, 4.74
V1222/B2816 (Wuhan-Hu-1) N=19					
n	19	17	17	19	19
GMT	109.83	448.98	3.63	391.02	3.56
95% CI	39.98, 301.71	215.26, 936.43	1.82, 7.25	178.75, 855.35	2.04, 6.21
V1222/B1222 (Beta) N=20					
n	20	18	18	19	19
GMT	96.52	306.77	3.10	274.21	3.01
95% CI	37.40, 249.15	125.71, 748.58	1.77, 5.42	124.11, 605.85	1.72, 5.27
V1222/B2816 (Beta) N=19					
n	19	17	17	19	19
GMT	65.82	544.07	7.19	515.81	7.84
95% CI	27.41, 158.08	310.44, 953.52	3.91, 13.19	256.36, 1037.84	4.41, 13.91

Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ 787,339) are imputed at the ULoQ value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
B1222 or B2816, Participants receiving a third dose booster of AZD1222 or AZD2816; CI, Confidence interval; GMT, Geometric mean titre; GMFR, Geometric mean fold rise; LLoQ, Lower limit of quantification; n, Number of subjects in analysis; N, Number of subjects per treatment group; ULoQ, Upper limit of quantification; V1222, Participants previously vaccinated with 2 doses of AZD1222
Derived from: Table 14.2.1.2

Additional Supportive Literature for AZD1222 as a Homologous and Heterologous Booster Dose.

Additional available clinical evidence (immunogenicity and safety) that supports the use of an AZD1222 third dose as a homologous and heterologous booster comes from the following:

- COV001 substudy conducted by University of Oxford that assessed AZD1222 as third dose homologous booster in participants who had previously received a 2-dose primary vaccination with AZD1222 (Flaxman et al 2021, previously submitted).
- COV-BOOST study conducted by University Hospital Southampton NHS that assessed an AZD1222 booster dose following a primary 2-dose vaccination with AZD1222 or BNT162b2 (Munro et al 2021).
- CoronaVac study in Brazil that compared immunogenicity of heterologous and homologous booster dosing with 4 vaccines, including AZD1222, in individuals who were previously vaccinated with a primary series of CoronaVac vaccine (Clemens et al 2022).
- A study conducted by University of Oxford using sera collected from individuals who had received 3 doses of AZD1222, which showed that a booster dose of AZD1222 significantly increased levels of antibodies against the Omicron variant (Dejnirattisai et al 2022).
- Results from analysis of sera from participants in Study D7220C00001 who had received 3 doses of AZD1222 for neutralising activity against Omicron variant in collaboration with the University of Oxford researchers and the UK Health Securities Agency (UKHSA, formerly called Public Health England).

Relevant results from these studies are summarised below.

Evidence for AZD1222 as a Homologous Booster; Substudy of COV001.

A substudy in 90 participants in the University of Oxford-sponsored study COV001 (Flaxman et al 2021) provided the initial data on the immunogenicity and reactogenicity of a third-dose booster of AZD1222 following a primary course of AZD1222/AZD1222. In this study, a third-dose booster of AZD1222 administered 28 to 38 weeks after the second dose induced high levels of antibodies, including against the Beta and Delta variants, and was responsible for maintaining Spike-specific T cell responses. This study was not randomised.

Antibody levels after the third dose were significantly higher than after the second dose. The median Spike IgG titre was 1792 ELISA units (IQR 899-4634) at 28 days after the second dose versus 3746 ELISA units (IQR 2047-6420) 28 days after the third dose.

Neutralising antibody titres after a third dose, measured in a randomly selected sub-population, were higher than those after the second dose against the Alpha ($p = 0.0023$), Beta ($p < 0.0001$), and Delta ($p < 0.0001$) variants.

Conclusion

The authors conclude that a third AZD1222 dose results in a further increase in immune responses, including increased neutralisation of variant SARS-CoV-2 viruses and could be used to increase vaccine efficacy against variants in susceptible populations. A booster of AZD1222 administered 28 to 38 weeks after the second dose was well tolerated by participants. Reactogenicity was consistent with the known reactogenicity profile of AZD1222, with fewer reactogenic events following a booster than after the first dose.

Evidence for AZD1222 as a Heterologous Booster; COV-BOOST and Com-COV Studies Study Design.

COV-BOOST was a multicentre, randomised, controlled, Phase II study of third-dose booster vaccination against COVID-19 in patients that had been previously vaccinated with 2 doses of AZD1222 or BNTT162b2 (Pfizer-BioNtech) (Munro et al 2021). The study was sponsored by the University Hospital Southampton NHS Foundation Trust and conducted outside of the AstraZeneca-Oxford collaboration. Participants were over 30 years of age and had no history of laboratory-confirmed SARS CoV-2 infection. At least 70 days had passed after the second of 2 doses of AZD1222 or at least 84 days after the second of 2 doses of BNT162b2 (Pfizer BioNtech). Coprimary outcomes were safety and reactogenicity and immunogenicity of anti-Spike IgG measured by ELISA.

In the COV-BOOST study, the geometric mean ratio was calculated by comparing post-booster GMTs to the corresponding value in a meningococcal vaccine control group. In addition, it is important to compare the neutralising antibody GMT elicited by a booster dose of a vaccine with the neutralising antibody GMT elicited after the primary series. As the post-primary series GMTs were not available in COV-BOOST study, the post-booster GMTs in the COV-BOOST groups are compared here to the post-primary series GMTs reported in the Com-COV study.

Com-COV was a randomised, controlled non-inferiority study conducted at 8 centres in the UK to investigate the safety and immunogenicity of heterologous versus homologous primary vaccination series (Liu et al 2021). A total of 463 participants, with a mean age of 57.8 years with a 28-day prime-boost interval were included in this analysis. Participants had no or well controlled comorbidities and had no history of laboratory-confirmed SARS CoV-2 infection.

The most relevant results of the COV-BOOST trail in the context of this variation are summarized in Table 29, which shows the results obtained in terms of anti-Spike IgGs, pseudoneutralizing and live virus neutralizing antibodies. Besides, comparison on the antibody titers observed in trial Com-COV as compared to those observed in the COV-BOOST study are shown in Table 30.

Table 29 Immune responses by third dose vaccine allocation and priming vaccine schedule 28 days after boost dose among the COVID-19 naïve modified intention-to-treat population

	Group A, AZD1222 primary course		Group A, BNT162b2 primary course	
	Prime with AZD1222/AZD1222 Control (n=93)	Prime with AZD1222/AZD1222 AZD1222 (n=100)	Prime with BNT162b2/ BNT162b2 Control (n = 111)	Prime with BNT162b2/ BNT162b2 AZD1222 (n = 98)
SARS-CoV-2 anti-spike IgG, ELU/mL				
Day 28	801 (664–967; n=91)	2457 (2058–2933; n=99)	2541 (2110–3060; n=111)	13424 (11702–15399; n=97)
Pseudotype virus neutralising antibody (wild-type), normalised 50% neutralising antibody titre				
Day 28	84.9 (68.7–105.0; n=90)	193 (161–231; n=98)	157 (129–192; n=111)	950 (802–1126; n=98)
Pseudotype virus neutralising antibody (Delta), normalised 50% neutralising antibody titre				
Day 28	20.0 (15.6–25.7; n=91)	48.9 (39.7–60.2; n=99)	37.9 (30.5–47.1; n=111)	260 (217–313; n=98)
Live virus neutralising antibody, normalised 80% neutralising antibody titre				
Day 28	146 (111–191; n=32)	346 (263–454; n=31)	531 (377–748; n=38)	2614 (2075–3294; n=40)

Data are geometric mean (95% confidence interval; number of samples available).

Derived from: Munro et al 2021, Table 5.

Table 30 SARS-CoV-2 Anti-spike IgG and pseudotype neutralising antibody at Day 29 following a primary vaccination series in the Com-COV study and following a primary vaccination series and an AZD1222 booster in the COV-BOOST study

Assay	Statistic	AZD1222 / AZD1222 (Com-COV)	AZD1222 / AZD1222 + AZD1222 (COV-BOOST)	BNT162b2 / BNT162b2 (Com-COV)	BNT162b2 / BNT162b2 + AZD1222 (COV-BOOST)
SARS-CoV-2 anti-spike IgG	n	105	99	110	97
	GMC (95% CI) in ELU/mL	1387 (1186-1623)	2457 (2058-2933)	13938 (12358-15719)	13424 (11702-15399)
Pseudotype neutralising antibody	n	101	98	102	98
	GMT (95% CI) in NT_{50}	61 (50-73)	193 (161-231)	574 (475-694)	950 (802-1126)

CI, Confidence interval; GMC, Geometric mean concentration; GMT, Geometric mean titre; ELU, ELISA laboratory unit; NT_{50}, 50% neutralising antibody titre.

Derived from: Liu et al 2021 (the Com-COV study) and Munro et al 2021 (the COV-BOOST study).

Conclusions

The authors concluded that all vaccines studied boosted antibody and neutralising responses after an initial course of AZD1222/AZD1222, with no safety concerns, and that the substantial differences in humoral and cellular responses in combination with vaccine availability will influence policy choices for booster vaccination. In addition, heterologous boosting with AZD1222 on top of an initial course of an mRNA vaccine (BNT162b2/BNT162b2) showed similar results as after an initial course of AZD1222/AZD1222 followed by a booster, with no safety concerns.

Given that participants in the COV-BOOST study were older than those in the Com-COV study, AstraZeneca hypothesizes that the COV-BOOST participants would have a diminished immune response compared with the Com-COV participants. It is therefore notable that SARS-CoV-2 anti-spike IgG and

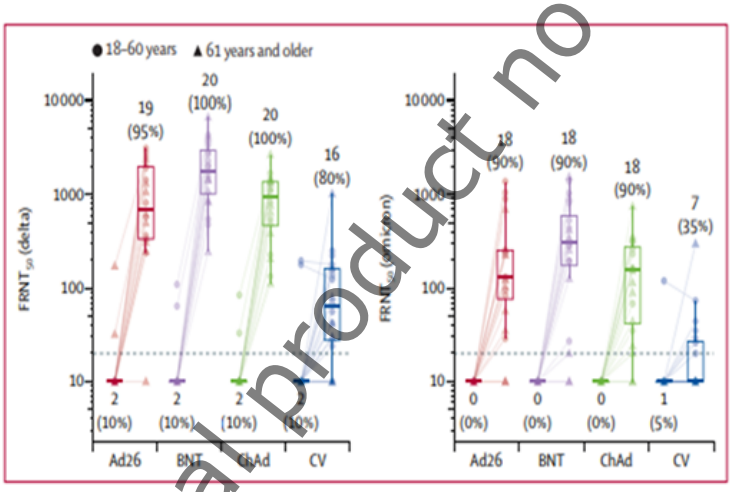
pseudotype neutralising antibody titres in participants who had received an AZD1222 booster still exceeded the corresponding titres in the younger participants receiving the BNT162b2/BNT162b2 primary series in Com-COV.

These data, in combination with safety and immunogenicity data from Study D7220C00001, are sufficient to demonstrate the benefit of AZD1222 as a heterologous booster dose. *Evidence for AZD1222 as a Heterologous Booster; RHH-001 Study (CoronaVac Study) Study Design*

This is a phase IV randomised single-blind study conducted in Brazil among 1240 participants 18 years or older to assess the safety and immunogenicity of a third heterologous booster dose of AZD1222, BNT162b2, and AD26.COV2-S compared to a third dose homologous booster of CoronaVac in adults previously vaccinated with a primary series of CoronaVac (Clemens et al 2022). The primary outcome measure was anti-Spike antibody titre at 28 days following booster dose. Secondary outcome measures were pseudoneutralising antibody titre and safety measures, including reactogenicity. Non-inferiority of heterologous schedules to homologous schedule was tested using a non-inferiority margin of 0.67 for the geometric mean ratio (heterologous vs homologous) of anti-spike antibodies following 28 days after the booster doses. A subset of 80 participants (20 per group, stratified by age) were also tested for live virus neutralisation using Delta and Omicron variants.

The geometric mean titres were increased following a booster across all groups with substantially higher seropositive rate (at least 90%) in heterologous booster groups, including AZD1222, as compared to that observed in the homologous booster group (35%) (Figure 7).

Figure 7 Live virus neutralisation titres against delta and omicron variants before and after 28 days following booster vaccination by booster vaccination groups



In each group, ten samples were selected from each age group (18–60 years, 61 years and older). Lines connect values from the same participant. Dotted line shows lower limit of the assay. Values below the limit were substituted with a titre of 10. Participants with antibody titres above the lower limit are considered seropositive and are shown as percentages. The midlines of the boxes show medians and the outer bounds of the boxes show IQRs. Error bars extend to the last data point within 1·5 ×the IQR above or below the 75th or 25th percentile.

4.3. Discussion

Introduction

The purpose of this variation is to support the use of AZD1222 as COVID-19 vaccination booster dose in adults 18 years and older, previously vaccinated with primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based). Consequently, an update of several sections of the SmPC is proposed. The main supporting data derive from Study D7220C00001.

Study D7220C00001 is an ongoing trial that was originally designed to evaluate the safety and immunogenicity of AZD2816 (a modified AZD1222 vaccine targeted against the Beta variant of SARS-CoV-2) as a 1-dose booster vaccination in previously vaccinated adult participants and as a 2-dose primary vaccination in previously unvaccinated adult participants. This study is also investigating the safety and immunogenicity of (1) a 2-dose vaccination with AZD1222 as first dose and AZD2816 as the second dose and (2) a 1-dose booster of AZD1222 in participants previously vaccinated with a 2-dose COVID-19 vaccine (either AZD1222 or an mRNA vaccine). The booster dose of AZD1222 was to be administered at least 90 days after the second dose of AZD1222 or mRNA vaccine.

The MAH is not seeking an indication for the product AZD2816 at this time due to its limited relevance in an epidemiological setting dominated by Delta and Omicron variants. Moreover, data from the previously unvaccinated cohort, who are to receive a 2-dose primary series of AZD1222 and/or AZD2816, have not been submitted by the MAH in the context of this variation.

Therefore, the data submitted here by the MAH (which include an Interim CSR and a clinical overview) are primarily focused on data from the AZD1222 booster treatment arm (subjects that previously received two doses of AZD1222 or an mRNA vaccine) in participants who were SARS-CoV-2 seronegative at study start. It is noted that this interim analysis includes a full analysis of the booster treatment group through Day 29 following AZD1222 booster. In agreement with the MAH, the assessors consider that these data could provide sufficient information regarding the immunogenicity and safety of the AZD1222 booster dose.

Conventions

Within the previously vaccinated cohort there are 2 sub cohorts, referred to as the 'AZD1222 cohort' (ie, those who previously received 2 doses of AZD1222) and the 'mRNA cohort' (ie, those who previously received 2 doses of a COVID-19 mRNA vaccine). Treatment groups are identified by 'V1222' and 'VmRNA', which refer to the cohorts based on their pre-study primary 2-dose course of vaccination, and by 'B1222' and 'B2816', which identify the booster dose received by that treatment group during the study. For example, V1222/B2816 refers to participants previously vaccinated with 2 doses of AZD1222 who received a Booster dose of AZD2816.

Regulatory History

The design of the CT D7220C00001 has been discussed in two Scientific Advice procedures plus and additional query (on historical control) that were posed by the MAH to the CHMP.

Some of the recommendations made by the CHMP in the final advice letters (FALs) have been followed, such as the non-inferiority analysis for GMT ratio (primary endpoint) and the seroresponse rate (key secondary endpoint), and the request to separate the original SAP into 3 individual SAPs with one specific to the AZD1222 previously vaccinated cohort, and another one to the mRNA previously vaccinated cohort. However, some CHMP recommendations were not followed by the MAH. In particular:

-i) Based on the fact that the percentage of seroresponders post-second AZD1222 dose (with a 4-week dosing interval) was higher (96.8%) when using live neutralization than when using a pseudoneutralization assay (59.7%), the CHMP recommended for immunogenicity comparisons to use preferentially the wild-type virus neutralization assay. Nonetheless, the CHMP considered acceptable to use of a pseudovirus neutralisation assay if adequate correlation between the assays was demonstrated. The MAH has provided only pseudovirus neutralising data and thus the MAH was asked to provide this report showing high correlation between the two assays. As detailed in section 9. , the MAH has provided new data from an analysis of concordance between the live virus neutralisation and pseudoneutralisation assays for the Wuhan strain in a population of participants boosted with AZD1222 from trial D7220C00001. The data show good agreement of the two assays in terms of geometric mean fold rises

and seroresponse rates. Thus, the use of the pseudovirus neutralisation assay in the trial D7220C00001 for the assessment of nAb responses after AZD1222 booster is considered justified.

-ii) The CHMP indicated that in order to include a claim in the product information so that AZD1222 can be used to boost the response in persons previously vaccinated with COVID-19 mRNA vaccines, the comparisons to be made were: "*… the company should plan a head-to-head comparison in which the response following a AZD1222 or AZD2816 booster is demonstrated to be non-inferior to the response with an mRNA booster. Alternatively, the response 28 days following the AZD1222 or AZD2816 booster response is demonstrated to be non-inferior to the response 28 days after the two primary doses of mRNA vaccine.*" Instead of following the CHMP advice, the MAH compared the response following AZD1222 booster to a historical control group that received two doses of AZD1222. The MAH's rationale is that immunobridging of neutralising antibody data in participants that received an heterologous booster to the historical control group allows the comparison of neutralising antibody titres post-boost to a regimen that is shown to be clinically protective after a primary series. A question was asked to the MAH to further justify not following the CHMP advice (see sections 7. 9. . In the response, the MAH followed the same original rationale and further discussed the data from the COM-CoV and COV-BOOST studies in support of deviation from the CHMP recommendation (see below for a detailed discussion on the data from the COV-BOOST study). As detailed in section 12., the MAH indicates that it was not possible to access serum samples for the VmRNA cohort nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study. As further discussed below, it is considered that despite not following the CHMP advice, the new data submitted during this procedure supports the use of AZD1222 as a heterologous booster.

It is noted that the original intention of CT D7220C00001 was to use the unvaccinated cohort that would receive two doses of AZD1222 as a comparator for the vaccinated cohorts that received a booster dose of AZD1222. Due to the problems in recruiting unvaccinated individuals the MAH consulted with the CHMP on using an historical control group as the primary vaccination comparator for the booster treatment arms instead of the in-study primary vaccination cohort. The CHMP agreed that the primary and key secondary non-inferiority analyses would therefore compare the previously vaccinated participants that received a booster dose in this study (D7220C00001) 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, predominantly conducted in the US and South America.

Overall Study Design of CT D7220C00001

The trial intended to recruit 1300 previously vaccinated participants (700 seronegative subjects previously vaccinated with 2 doses of AZD1222 and 600 previously vaccinated with 2 doses of an mRNA vaccine) to receive an additional single-dose booster vaccination. In addition, seropositive participants were enrolled (with a cap of 10% of the seronegative population) to support exploratory analyses in these participants. The previously vaccinated cohorts (with either AZD1222 or an mRNA vaccine) were to be randomised 1:1 to 1 dose of AZD1222 or AZD2816. This booster dose cohort was double-blinded.

The treatments were: AZD1222 (nominal dose of 5×10^{10} viral particles), which corresponds to the EU authorized vaccine Vaxzevria, and AZD2816 (nominal dose of 5×10^{10} viral particles) that was selected to match with the approved Vaxzevria vaccine.

Immunogenicity was to 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. The inclusion and exclusion criteria are considered adequate, and overall, it is considered that the participants to be recruited represent the real-world population that would receive a booster dose of AZD1222. It was, however, allowed the inclusion of previously vaccinated participants with a 2-

dose primary vaccination with either AZD1222 (with a 4- to 12-week dosing interval) or with an mRNA vaccine (e.g., BNT162b2 vaccine [Pfizer-BioNTech] with a 3- to 12-week dosing interval or mRNA-1273 vaccine [Moderna] with a 4- to 12-week dosing interval). Allowing a wide dosing interval for primary vaccination could be a confounding factor when comparing the immune responses to the historical control group since participants from this group had a dose interval of 4 weeks.

It is noted that the claim in the SmPC indicates that the AZD1222 vaccine can be used in subjects 18 years of age and older. Considering that in immunological terms, subjects from 18 -29 years of age are comparable to young adults (older than 30 years), the lack of subjects from 18 -29 years of age in the trial does not question the use of the AZD1222 vaccine in subjects from 18 years of age.

Objectives and endpoints

As already mentioned above, following database lock, the CHMP requested changes to the primary and key secondary immunogenicity objectives as well as separating the original SAP into 3 individual SAPs with one specific to the AZD1222 previously vaccinated cohort, and another one specific to the mRNA previously vaccinated cohort.

Cohort of AZD1222 previously vaccinated subjects. 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. For GMT ratio, non-inferiority was demonstrated if the lower limit of the 2-sided 95% CI of the GMT ratio of the comparator group and the reference group is > 0.67.

Regarding the key secondary endpoint on the difference in seroresponse, non-inferiority was demonstrated if the lower bound of the 2- sided 95% CI rate difference in seroresponse between the comparator group and reference group was ≥-10%.

These two analyses will compare the previously vaccinated participants that received a booster dose in study D7220C00001 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.

The primary and key secondary endpoints as well as the non-inferiority criteria to be used are in line with those recommended by the CHMP in the SAs posed by the MAH. Similarly, the use of an historical control group for the comparisons was agreed by CHMP, as long as there was a good match with the cohort that received an AZD1222 booster cohort.

The other secondary (pseudoneutralizing titers against VOC Beta, spike-specific IgG response to SARS-CoV-2 and the anti-vector neutralizing antibody titres to the ChAdOx-1 adenovirus vector) and exploratory (GMTs and seroresponse to the Alpha, Beta, Gamma, and Delta variants and B-cell and T-cell responses) endpoints are considered adequate to better understand the immune responses induced after a booster dose.

The sensitivity analyses for the primary endpoint (including BMI and age as continuous covariates, as well as taking into account the dosing interval between the primary series vaccination for the previously vaccinated) are endorsed.

Cohort of mRNA previously vaccinated subjects. The primary and secondary immunogenicity endpoints used by the MAH are the same used for the AZD1222 previously vaccinated cohort and as mentioned above.

Assays used to assess SARS-CoV-2 immune response.

The immune response raised by the vaccine was analysed both in terms of the humoral (SARS-CoV-2 pseudovirus neutralizing antibodies -Wuhan and beta variant-, SARS-CoV-2 S- binding antibodies -Wuhan, alpha, beta and gamma variants-, and anti-ChAd0x1 neutralising antibodies) and cellular immune response (SARS-CoV-2 IFNγ ELISpot Assay). The overall approach is endorsed.

The different methods used are adequately described by the MAH and the most relevant assays are validated (Pseudovirus Neutralising Antibody Assay –Wuhan and beta variant- and the multiplexed ECL method for the detection of antibodies to SARS-CoV-2 antigens). The validation reports are considered adequate to support the good performance of the different assays. The assays to test the neutralizing antibody response to the Delta and Omicron variants were not validated.

Randomisation and Blinding

Treatment was double-blinded for all previously vaccinated participants receiving a booster dose (either AZD1222 or AZB2816). Randomisation was made according to the treatment (AZD1222 or AZD2816) and stratified based on age (< 65, ≥ 65), sex, and presence of comorbidities, which is considered adequate.

It is noted that the randomisation made in the trial (based on the vaccine received -either AZD1222 or AZB2816-) makes sense for the immunogenicity comparisons between these two vaccine treatments. However, the randomisation (and stratification) made is not relevant for comparing the immunogenicity response of the AZD1222 boosted cohort with that of historical control group (that received 2 doses of AZD1222).

Statistical Methods. As already mentioned above the MAH has followed the request from CHMP with reference to provide independent SAPs for the analysis of the two booster cohorts. Moreover, the non-inferiority comparisons based on the GMTR and the difference in seroresponse as well as the non-inferiority margins for the analysis of the AZD1222 booster dose in subjects that previously received two doses of AZD1222, were in agreement with the CHMP advice. This was not the case for those previously vaccinated with an mRNA vaccine, since CHMP advice was not followed.

The analyses of GMT/GMR were performed based on a model-adjusted titre level. The model adjusted analyses were derived using analysis of covariance (ANCOVA) models that included the log transformed value of the titre or result as the dependent variable. This approach is endorsed. The MAH has considered an historical control group for testing the primary and a number of secondary endpoints. Selection of the historic control group occurred before database lock and the unblinding of data. This historical group is based on subjects previously vaccinated with two doses of AZD1222 in study D8110C00001.

A one-to-one propensity score matching was used to match the studies D8110C00001 and D7220C00001 based on the following covariates: age, gender, BMI, and presence of baseline comorbidities. Likewise, a caliper was considered to ensure a common support region for the covariates from both studies. The Mahalanobis distance derived from the logit of the propensity score adjusting for BMI and age has been applied. In the current setting, the use of the historical control is understood and acceptable for the purpose of the current study. However, the acceptability of this external cohort is strongly conditioned on the Applicant's proper design and implementation at all stages of the procedure. This is due to the inherent biases and known/unknown confounders of an incorporation of a non-randomised arm, which might hamper the overall interpretation of the analyses. For these reasons, a potential lack of robustness should be accompanied with appropriate justifications and discussions on the impact on the interpretation of the results, which has not been provided in detail. The MAH was asked to provide key baseline comparability to assess the similarity of the subjects previously vaccinated with two doses of AZD1222 from the study D8110C00001 and the subjects recruited in study D7220C00001, and therefore a comparative table with data of these differences was requested to the MAH. As detailed in section 9. , the MAH has provided the requested information.

It is considered adequate that the population for the primary and secondary immunogenicity analysis was the "Seronegative Immunogenicity Analysis Set", that included all randomized participants (seronegative at baseline) who received a booster dose of AZD1222, had baseline and post-dose antibody measurements, and had no protocol deviations to interfere with interpretation of the antibody response data. Moreover, comparisons of antibody titres between the previously vaccinated cohort in study D7220C00001 and the historical controls from Study D8110C00001 were conducted using the Seronegative Immunogenicity Analysis Set.

The MAH was asked if the sample size was updated accordingly once the three independent SAPs were prepared (see section 10.). The MAH acknowledged that the sample size was not updated since the data were already locked, and enrolment had completed. The MAH has clarified that the sample sizes within each sub-SAP provide adequate power for hypothesis testing. This was considered acceptable, and the issue was considered solved.

A hierarchical multiple testing has been performed to control the type I error across different comparisons in a sequential order. This strategy is considered acceptable from a statistical point of view.

While the comparative analyses based on the interim analysis dataset will be repeated at the primary analysis (previously the first-time comparative analyses were planned to be conducted), it is noted that the interim analysis dataset included all previously vaccinated seronegative participants randomised to the study and that bioanalysis of baseline and Day 29 pseudoneutralising antibodies was complete for all of these participants at database lock. Consequently, as the results presented in the Interim CSR are not anticipated to change when using the primary analysis dataset, hypothesis testing and conclusive assessment could be conducted for the key endpoints.

Changes in the Conduct of the Study or Planned Analyses

In total three protocol amendments were implemented. The first one was implemented before participant recruitment and implied that all participants in the AZD1222 booster treatment groups were ≥ 30 years of age, in accordance with the UK-specific CSP amendment. The two latest ones to incorporate recommendations made by CHMP in the FAL to the two SAs and the query (regarding use of a historical control group) posed by the Company.

The MAH has now submitted an interim CSR, and the final analysis will occur when data from all vaccinated participants is available through completion of the last study visit at 180 days after the final dose of study intervention. It is noted that the immune response of AZD1222 against the Beta variant in the historical control group (from in study D8110C00001) are not yet available, and thus the comparisons of the response to the Beta variant will be reported in the Primary Analysis CSR.

RESULTS

Study participants

This study is being conducted at 35 sites in Brazil, Poland, South Africa, and the UK. It is noted, however, that previously vaccinated participants were recruited only from 19 study sites in the UK and 4 study sites in Poland. In total, 1379 participants received a booster dose of AZD2816 or AZD1222, which is in line with the intended figure of 1300 subjects stated in the original CT protocol.

It is pointed out that the data regarding binding antibodies were lacking for 27 subjects and also Day 29 data for 17 seropositive participants. Since primary and key secondary endpoints analysis were based on pseudoneutralising antibodies in baseline seronegative subjects it is concluded that the absence of these data does not affect the analysis of primary and key secondary immunogenicity endpoints.

The participants (previously vaccinated with two doses of AZD1222 or an mRNA vaccine) were randomized to receive one dose of AZD1222 or AZD2816, and the disposition of participants was

generally well balanced across the two treatments (AZD1222 or AZD2816). It is noted that in the context of this variation, in which the primary and secondary endpoints imply comparison with an historical control group, the relevant information is the adequate match of the AZD1222 boosted cohort with the cohort of participants from the historical control group.

In the mRNA cohort, all but one of the participants (that received mRNA-1273 vaccine) were vaccinated with two doses of BNT162b2.

The number of subjects (both seropositive and seronegative at baseline) previously vaccinated with an mRNA vaccine (322) was lower than those previously vaccinated with AZD1222 (367 subjects). Within each group all participants continued in the study at data cut-off. Important protocol deviations that were deemed to interfere with the immune response results affected few participants (only 6 participants, 3 in the V1222/B1222 and 3 in the VmRNA/B1222 groups). The seronegative immunogenicity set used for the primary and key secondary endpoints includes 342 participants from the V1222/B1222 and 294 participants from VmRNA/B1222 groups. The historical control group included 508 subjects. The corresponding figures for the seropositive immunogenicity set were 20 and 23 participants. It is noted that, for both booster cohorts, more that 98% of the randomized participants were included in the corresponding immunogenicity analysis sets.

Demographic and Other Participant Characteristics.

The demographic characteristics of the historical and the V1222/B1222 groups were similar in terms of age, sex and BMI.

It is noted that there were differences between the VmRNA/B1222 and the other two groups (historical control group and V1222/B1222). The median age of participants previously vaccinated with an mRNA vaccine (55 years of age) was lower than the median age of participants previously vaccinated with AZD1222 and the historical control group (62 years of age). The sex of the participants also differed between the cohorts, with over 60% of the mRNA cohort, 45% in AZD1222 and 47% in the historical control groups being female. The MAH explained that the observed differences in demographics and baseline characteristics between the two cohorts (V1222/B1222, VmRNA/B1222) were the result of vaccination rollout timeline in the UK. This explanation is well justified by the MAH. However, this explanation does not justify using a historical control group with different characteristics than the VmRNA/B1222 cohorts for immunogenicity comparisons. The V1222/B1222 cohort includes a significant proportion of subjects older than 65 years of age (45.9%).

In order to get a clear picture of these three populations, the MAH was asked to provide a head-to-head comparison of the demographic and baseline characteristics of the V1222/B1222, VmRNA/B1222 and the historical control group. As detailed in section 13, the MAH has provided all the requested information. The MAH explained that race/ethnicity was not expected to impact the efficacy, immunogenicity, or safety of AZD1222 and, as such, ethnicity was not selected as matching criteria. This approach is agreed upon and thus the differences observed in race/ethnicity between the historical control group and the booster cohorts are not expected to have an impact on the immunogenicity comparisons made. The MAH also discussed the implications of the differences in several characteristics (age, sex) between the VmRNA/B1222 and V1222/B1222 (and historical control group) groups. The MAH concluded (based on the fact that the results of the model-adjusted analyses were consistent with the raw data results and the subgroup analyses of the immunogenicity) that these differences do not have a clinically meaningful impact on the primary and secondary immunogenicity analyses.

It is also noted that the median time since previous vaccination (i.e., second dose of AZD1222 or mRNA vaccine) was approximately 9 months in the AZD1222 cohort (range: 2.5 months to > 1 year), and 4 months in the mRNA cohort (range: 2.5 to 7 months). The MAH proposes the following text for section 4.2 of the SmPC: "The third dose should be administered at least 3 months (90 days) after completing

the primary vaccination course." The MAH was asked to justify this period of time (3 months). As detailed in section 13, the immunogenicity results obtained from a significant proportion of subjects who received an AZD1222 booster with a dosing interval of 2.5 to 6 months from primary vaccination provides support for the claim in the SmPC.

It is also noted the important differences in the primary vaccination dosing interval in the different groups to be analysed. The median time between the first two primary vaccination doses was 59 days for the V1222/B1222 group, 70 days for the VmRNA/B1222 group, and the period for the historical control group has not been provided (although it is expected to be around 29 days based on previously submitted data from trial D8110C00001). This information from the historical control group was requested to the MAH. As detailed in section 13, it is now clearly stated, that the median primary vaccination dosing interval in the historical control group was 28 days. In addition, the differences in the primary dosing interval do not appear to have a significant impact on the immunogenicity comparisons made

There were 13 seronegative participants who received the booster dose ≥ 70 days but < 90 days after their second dose. The MAH decided, prior to database lock, to include these participants in the Seronegative Immunogenicity Analysis Set. The MAH was asked to to justify including in the analysis participants with less than 90 days from second dose to booster dose. The MAH provided data which showed that the incorporation of subjects who received the booster dose 70 days or more but fewer than 90 days after their second dose do not show any significant impact in the immunogenicity results obtained.

In conclusion, after assessment of the responses to the RSI m it is concluded that adequate information has been submitted regarding the baseline and demographic characteristics for the historical control group. Moreover, although some of the characteristics of historical control group were not well matched with those of the V1222/B1222 and the VmRNA/B1222 groups, the MAH has provided adequate data to exclude that these differences had a meaningful impact on the immunogenicity comparisons made.

Immunogenicity Results for AZD1222 Booster Dose in the Previously Vaccinated Cohorts (Primary Series of AZD1222 or an mRNA vaccine)

AZD1222 Cohort (AZD1222 as a Homologous Booster)

In the AZD1222 cohort, baseline neutralising antibody titres, measured before administration of the booster, were below the LLoQ in approximately 75% of participants as against the Wuhan-Hu-1 strain and approximately 87% as against the Beta variant.

At Day 29 after a booster dose of AZD1222 (in the previously AZD1222 vaccinated cohort), an increase in GMTs (pseudoneutralising antibodies) against both Wuhan-Hu-1 strain and Beta variant was observed, representing a 6.54 and 7.63 fold-rise over baseline level, respectively.

The pseudoneutralising antibody seroresponse (ie, ≥ 4-fold increase in titres from baseline) to a booster dose of AZD1222 against Wuhan and Beta strain resulted in lower seroresponse rates of 66% in both cases.

The primary immunogenicity endpoint was met since the Wuhan-Hu-1 pseudoneutralising antibody GMT ratio at Day 29 after AZD1222 booster to that at Day 29 after dose 2 of primary series in historical control was 1.03 (95% CI 0.917, 1.146), a result that fulfilled the 1.5-fold non-inferiority criterion (ie, lower bound of the 2-sided 95% CI for GMTR >0.67). It is noted that the GMTR figure obtained (1.03) indicates that the booster dose does not increase the antibody titres reached after two AZD1222 doses.

However, the pre-specified non-inferiority criterion for difference in seroresponse rate against Wuhan-Hu-1 between the group that received the AZD1222 booster and the historical control was not met (lower bound of 95% CI = −24.0%) since the lower bound of the 2- sided 95% CI rate difference had to be ≥-

10%. It is noted that the difference in the seroresponse rate between the two groups was 18%, corresponding to a rate of 84.1% in the historical control and 66.1% in the participants that received a booster of AZD1222. It is noted that the CHMP stated in the FAL (EMA/SA/0000073209; 7/December/21): "*it would be expected that all or very nearly all subjects will have at least a 4-fold increase in neutralizing antibody after the third doses. The seroresponse rates can be designated secondary and the company should appreciate that any result that is less than ~95% will be of concern in a primed population.*" Thus, the percentage of seroresponders observed is considered very low (61%) and this observation questions the indication sought for a booster AZD1222 dose. The MAH justifies not meeting this key secondary endpoint on the basis that the baseline GMT against Wuhan-Hu-1 strain were approximately twice as high in participants from AZD1222 cohort as compared to the historical controls (38.23 vs 20.01, respectively). In this situation, the MAH explains that naïve participants with lower (or non-existent) neutralising antibody titres at baseline are more likely to achieve a 4-fold rise in neutralising antibody titres post-booster. Although it is known that the baseline titres in fact have an impact on the seroconversion rates, a question was raised to the MAH to discuss the possibility that only a few numbers of high-responders to the booster vaccine are driving compliance with the non-inferiority of the primary endpoint based on the ratio of GMTs and this would result in failing the key secondary endpoint. Moreover, taking into account that baseline neutralising antibody titres (measured before administration of the booster dose) were below the LLoQ in approximately 75% of participants (against Wuhan-Hu-1 strain) the MAH is asked to determine the seroresponse rate in this subgroup in order to shed light on the booster effect of AZD1222. Moreover, considering the wide dosing intervals between the two primary vaccination doses and also between second and booster doses (described above), the MAH was asked to perform subgroup analysis to determine the impact of the dose interval in primary vaccination as well as the time interval from second to booster dose on the GMT and seroreponse rate reached.

As detailed in section 13, the MAH has provided adequate responses to these requests. In particular, the MAH provided histograms [(showing the percentage of subjects reaching different antibody titres (pre- and post- booster)], reverse cumulative distribution curves (RCDC) of pseudoneutralising antibodies (pre- and post- booster doses), and a specific analysis of the booster responses in participants with antibody tires pre-booster below the LLOQ. Overall, the histogram data and the RCDC curves for both the V1222/B1222 and the VmRNA/B1222 cohorts do not indicate that there are two different subpopulations, one of high responders and the other responding poorly to the booster dose. Rather, the data suggest that most of the subjects that received a booster dose increased the nAb titres. For both the V1222 and VmRNA groups, at day 29 after booster doses, a higher rate of seroresponse (81.4% and 97.3%, respectively) was observed in participants with baseline nAb titres < LLOQ than in the overall population [66.1% (V1222) and 43.2% (VmRNA)]. It is noted that in the historical control group the seroresponse rate after primary vaccination was 84.1%. Thus, the data show that AZD1222 booster injection strongly boosts the immune response in subjects from the VmRNA and V1222 groups with titres below the LLOQ. The lower seroresponse observed in the overall population from both groups is then a consequence of the difficulty of achieving a $\geqslant$ 4-fold rise in nAb titres in subjects with high titres before receiving the booster dose. In conclusion, the data provided do not indicate that meeting the primary endpoint (based on GMTs ratio) for both the V1222/B1222 and VmRNA/B1222 were due to a small number of high-responding study participants. Rather, the data indicate that most of the subjects increased the nAb titres after AZD1222 booster, although high seroconversion rates were observed in subjects with titres pre-booster bellow the LLOQ since it was more difficult to achieve seroresponse of $\geqslant$ 4-fold rise from baseline in subjects with higher pre-booster titres. This is a general observation made also for many other vaccines. In conclusion, although the seroresponse rates in the overall population from both boosted cohorts were far from the 95% figure indicated by the CHMP; it is considered that the high seroresponse rates seen in participants with baseline nAb titres < LLOQ in fact demonstrate the adequate boosting ability of AZD1222 dose. The MAH has provided adequate evidence showing that

neither the dosing interval in primary vaccination nor the time interval from second to booster dose have a meaningful impact on the immunogenicity comparisons made.

It should be noted that important differences in seroresponse rate were also shown when data from binding antibodies were analysed. In fact, after two doses of AZD1222, the seroconversion rate against the Wuhan strain in the historical control group was 98.8% whereas the seroconversion rate was only 68.2% for the group V1222/B1222. It is also mentioned that, according to S-binding antibodies, the seroconversion rate, 29 days after AZD1222 booster, against beta variant was also low (71%). Based on the good precision of the assay for detection of S-binding antibodies, the MAH is asked to submit and discuss the data on seroconversion (based on a ≥2-fold increase) both for the V1222/B1222 and the historical control group). As detailed in section 9. , the MAH calculated the seroresponse rates based on a ⩾ 2 fold rise in Spike-binding antibodies. The seroresponse rates determined when using a ⩾ 2 fold rise in binding antibodies were 82.6% (95%CI 78- 86), and 71.1% (95%CI 65-76) for the V1222/B1222 and the VmRNA/B1222 cohorts, respectively. These figures significantly increased from those calculated when using a ⩾ 4 fold rise in binding antibodies: 68.2% (95%C 62-73) for the V1222/B1222 and 36.7% (95%CI 30-42) for the VmRNA/B1222. The seroresponse rate was practically unaltered for the Historical control group 98.8% [(95%CI 97-99) to 99.2% (95%CI 98-99)].

The results based on a ⩾ 2 fold rise in binding antibodies have also been described in other studies aimed at measuring the immunogenicity reached following a homologous or heterologous boost with COVID-19 vaccines (e.g., Atmar et al. DMID 21-0012 Study Group. N. Engl. J. Med. 2022; 386:1046-1057). In the context of this trial, the data obtained are interpreted in that the seroresponse rate in terms of S-binding antibodies (⩾ 2-fold rise) is quite significant for both cohorts (V1222/B1222 and VmRNA/B1222), and thus these data indicate that the AZD1222 booster dose is in fact boosting the response induced after primary vaccination in most of the participants in the trial.

Certainly, the seroresponse rates reached are still lower than those achieved after primary vaccination series in the historical control group. It is also noted, that the relevance of S-binding antibodies for clinical protection is unclear, but a number of studies have shown that S- binding and nAb titres correlate, and thus it is expected an increase in protection in subjects with only a 2-fold increase in S-binding antibodies. In summary, the data described here together with those based on seroresponse rates of nAb in subjects with antibody titres below the LLOQ before booster dose (see query 1b) are interpreted in that the AZD1222 booster dose increases nAb antibody titres in a large proportion of the subjects receiving the AZD1222 booster. This interpretation was not so obvious when using seroresponse rates based on nAb in the overall populations of the two cohorts or when using on a ⩾ 4-fold rise of S-Binding antibodies.

In conclusion, all the data described above provide support for the use of AZD1222 as a homologous booster.

When measured according to the spike-binding antibody titres, the booster AZD1222 dose increased the GMT by 9.43, 10.29, 10.09, and 10.05-fold from baseline titres against the Wuhan-Hu-1, Beta, Alpha and Gamma variants, respectively. It is noted that the titres reached against these variants in the historical group are not available yet, and thus it is not possible to assess the increase of antibody titres against the different variants observed after the booster dose in comparison with that reached after a primary vaccination series.

mRNA Cohort (AZD1222 as a Heterologous Booster)

As already discussed above, the MAH did not follow the CHMP recommendation regarding the primary and key secondary endpoints to be analysed in order to get an indication for boosting of subjects that had received a primary vaccination with an mRNA vaccine. Instead, the MAH followed the immunobridging strategy followed for the analysis of the AZD1222 booster (i.e, comparison of immune

response of the VmRNA /B1222 group to the historical control group already used for comparison of the AZD1222 booster). The MAH's rationale is that immunobridging of neutralising antibody data in participants heterologously boosted to the historical control group allows the comparison of neutralising antibody titres post-boost to a regimen that is shown to be clinically protective after a primary series. A question was asked to the MAH to further justify not following the CHMP advice (see section 7. and section 9.). In the response to the preliminary questions, the MAH followed the same original rationale and further discussed the data from the COM-CoV and COV-BOOST studies in support of deviation from the CHMP recommendation (see below for a detailed discussion on the data from the COV-BOOST study). As described, in section 9. , the MAH indicates that it was not possible to access serum samples for the VmRNA cohort nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study. As further discussed below, it is considered that despite not following the CHMP advice, the new data submitted in the responses to RSI support the use of AZD1222 as a heterologous booster.

In the mRNA cohort, baseline neutralising antibody titres, measured before administration of the booster, were below the LLoQ in approximately 14% of participants as against the Wuhan-Hu-1 strain and approximately 30% as against the Beta variant.

At Day 29 after the AZD1222 booster (in the previously mRNA vaccinated cohort), an increase in the pseudoneutralising antibody titres against Wuhan-Hu-1 strain and Beta variant was observed, representing a 3.77 and 5.93-fold-rise in neutralization over baseline against these variants.

The proportion of participants in the VmRNA/B1222 group with a pseudoneutralising antibody seroresponse was low against the Wuhan-Hu-1 strain (43.2%) as well as against Beta (57.6%).

It is noted that at baseline (before receiving the booster dose) the VmRNA/B1222 cohort had considerably high GMT values [197 (95% CI 179-271)] that compared well to the GMT reached after primary vaccination in subjects from the historical control group [242 (95% CI 224-262)]. Considering the small difference between these GMT figures, reaching the primary endpoint proposed by the MAH is not a demanding requirement. The Wuhan-Hu-1 pseudoneutralising antibody GMT ratio at Day 29 after AZD1222 booster in the mRNA cohort to that at Day 29 after dose 2 of primary series in historical control was 3.08 (95% CI 2.781, 3.405), which met the 1.5-fold non-inferiority criterion (ie, lower bound of the 2-sided 95% CI for GMTR >0.67) stablished by the MAH.

The pre-specified non-inferiority criterion for difference in seroresponse rate against Wuhan-Hu-1 was not met (lower bound of 95% CI = -47.3%). The MAH indicated that this result was attributed to the higher baseline GMT values in previously vaccinated participants as compared the matched unvaccinated controls (197.74 versus 20.01). The same comments made above for the AZD1222/B1222 cohort (on the impact of the primary vaccination dosing interval and the dose interval between second and booster dose on GMT reached, on the possibility that the results observed could be due to only a few number of high-responders and the calculation of seroresponders in those below the LLoQ at baseline) were asked to the MAH. As already discussed above (assessment of the results of the V1222/B1222 cohort) and in section 9. , the new data provided by the MAH do not indicate that there are two different subpopulations, one of high responders and the other responding poorly to the booster dose. Rather, the data suggest that most of the subjects that received a booster dose increased the nAb titres. Moreover, a high rate of seroresponse (97.3%) was observed in participants from the VmRNA cohort with baseline nAb titres < LLOQ as compared to those seen in the overall population (43.2%). It is considered that the high seroresponse rates seen in participants with baseline nAb titres < LLOQ in fact demonstrate the adequate boosting ability of AZD1222 dose, whereas the lower seroresponse in the overall population reflects the difficulty in achieving high seroresponse in subjects with high pre-booster titre.

It should be noted that low seroresponse rates were also shown when data from binding antibodies were

analysed. In fact, at Day 29 after the AZD1222 booster dose, S-protein binding antibody seroresponse were 36.7% to 31.1% against the Wuhan-Hu-1 and Beta strains, respectively. As already discussed above (assessment of the results from the V1222/B1222 cohort) andin section 9.), the new data provided by the MAH showed that the seroresponse rates determined when using the criterion of $\geqslant$ 2 fold rise in binding antibodies was 71.1% (95%CI 65-76) for the VmRNA/B1222 cohorts, a figure much higher to that obtained [36.7% (95%CI 30-42)] when using the criterion of $\geqslant$ 4 fold rise in binding antibodies. These results together with those based on seroresponse rates of nAb in subjects with antibody titres below the LLOQ before booster dose (see query 1b) are interpreted in that the AZD1222 booster dose increases nAb antibody titres in a large proportion of the subjects receiving the AZD1222 booster. This interpretation was not so obvious when using seroresponse rates based on nAb in the overall populations of the two cohorts or when using on a $\geqslant$ 4-fold rise of S-Binding antibodies.

In conclusion, all the data described above provide support for the use of AZD1222 as a heterologous boost.

When measured according to the spike-binding antibody titres, there were increases of 3.24, 3.02, 3.37, and 3.08-fold-rise from baseline for the Wuhan-Hu-1, Beta, Alpha and Gamma variants, respectively. It is noted that the titres reached against these variants in the historical group are not available yet, and thus it is not possible to assess the increase of antibody titres against the different variants observed after the booster dose in comparison with that reached after a primary vaccination series.

Other secondary and exploratory endpoints

At Day 29 after a booster dose of AZD1222, neutralising responses against Delta variant and Omicron variant showed an increase in titres from baseline, both in in the AZD1222 cohort and in the mRNA cohort. The anti-Omicron antibodies present in subjects boosted with AZD1222 (from study D7220C00001) who were previously vaccinated with either AZD1222 or an mRNA vaccine were also analysed. The sera analysed were very limited (about 30 subjects per cohort). The results from these assays showed an increase in titres against Omicron variant following the booster dose, although it is noted that the neutralizing antibody titres to the Omicron variant were the lowest observed of any evaluated variant to date. Considering that these assays were not validated and given the limited sample analysed, the results are taken as exploratory

Subgroup Results

Pre-specified subgroup analysis was made based on age, sex, and comorbidity following booster doses of AZD2816 or AZD1222 in both the V1222 and VmRNA seronegative cohorts. Although some numerical differences were noted across subgroups, overall, the responses were similar, and no apparent statistical difference was observed.

As discussed above, the MAH was asked to carry out a subgroup analysis to determine the impact on the GMT titres reached depending on the dose interval in primary vaccination as well as the time interval from second to booster dose. The MAH has submitted the data requested. The results shown (section 9) do not indicate any obvious effect of the primary series dose interval or the time elapsed from primary vaccination to booster doses on the immunogenicity analysis performed both regarding the homologous or heterologous booster.

Cell-Mediated Immunogenicity

T cell responses were assessed in an IFNγ ELISpot assay with peptides specific to the Wuhan- Hu-1 strain (Wuhan S1 + S2) in a very limited number of subjects (about 20 per cohort, either receiving AZD1222 or an mRNA vaccine). Practically no increase in Spike-specific IFNγ T Cell responses were

observed in subjects previously vaccinated with either AZD1222 or an mRNA vaccine following a booster dose of AZD1222. This result also points out to the poor immune responses achieved after AZD1222 booster vaccination.

Immunogenicity (Anti-Vector)

Seroresponse in terms of anti-vector pseudoneutralising antibodies was higher in the VmRNA/B1222 cohort (93.4%) than in the V1222/B1222 cohort (59.7%). The data submitted indicate that minimal correlation was observed between ChAdOx1 pseudoneutralising antibody titres and pseudoneutralising antibody responses to SARS-CoV-2. These data appear to rule out the possibility that anti-vector antibodies could be responsible of the low immune response reached upon receiving three doses of AZD1222.

Results for Seropositive Participants

The characterization of pseudoneutralising antibody data for AZD1222-cohort participants who were seropositive at baseline is very limited (based on approximately 20 subjects for both the V1222/B1222 and VmRNA/B1222 cohorts). A booster dose of AZD1222 increased pseudoneutralising antibody titres, which was approximately half of that observed in seronegative participants. It is also noted that in this population, pseudoneutralising seroresponders to the Wuhan strain in cohort V1222/B1222 were only 36.8%.

Additional Supportive Literature for AZD1222 as a Homologous and Heterologous Booster Dose.

The additional available clinical evidence (immunogenicity and safety) submitted by the MAH in order to support the use of an AZD1222 third dose as an homologous and heterologous booster comes from a number of studies, many of them with very few subjects to reach any robust conclusion. The most relevant data derives from the COV-BOOST study where different vaccines were used to boost subjects that had received primary vaccination with two doses of AZD1222 or BNT162b2. It is noted that in this study, the geometric mean titre achieved, 28 days after the booster dose, was compared to the corresponding value in a meningococcal vaccine control group and not to the GMT reached 28 days after primary vaccination.

Both in the AZD1222/AZD1222-primed individuals boosted with AZD1222, and in the BNT162b2/BNT162b2-primed individuals boosted with AZD1222, the geometric mean ratio compared with the control group (in terms of anti-Wuhan antibodies) was in the range of 2 to 6 depending on the antibody assay used. Overall, these results are broadly in line with those described for the GMT fold increase pre- and post- booster observed in study D7220C00001. It is noted however, that the GMTR (Wuhan strain) reached in the AZD1222/AZD1222-primed subjects after an BNT162b2 booster was much higher (more than 20-fold increase) as compared to an homologous AZD1222 booster. No data on seroresponse rates was included in the publication that described the COV-BOOST results (by Munro, et al).

The MAH also compares the titres reached, 29 days after last vaccination, in participants from Com-COV and COV-BOOST studies, and it concludes that after receiving a homologous AZD1222 booster dose, both spike binding as well as pseudoneutralising antibody responses were numerically higher after the AZD1222 booster in COV-BOOST than the titres observed in the primary series in Com-COV study. It is however noted, that similar binding antibody titres were observed in subjects that received two doses of BNT162b2 as compared to those primed with two doses of BNT162b2 and later boosted with AZD1222. Thus, the interpretation of these comparisons has to be taken with caution.

The MAH provides also data from a trial (RHH-001) which compares a heterologous booster dose of AZD1222 compared to a third homologous booster dose of CoronaVac in adults. These data are not discussed in this assessment report since Coronavac is not currently authorized in the EU. Thus, inclusion of data from this trial in the SmPC, as proposed by the MAH, is not accepted.

In summary, the primary endpoint based on calculating GMT ratios between the nAb antibody titres reached 29 days after AZD1222 booster dose and those achieved 29 days after primary vaccination (derived from the historical control group) were met for both cohorts (AZD1222 and mRNA primary vaccinated subjects). The key secondary endpoint comparing the nAb seroresponse rate after primary vaccination with that pre- and post- booster dose was not met for any of the two cohorts. The new data submitted by the MAH, demonstrate that these results on the seroresponse endpoint were not due to a subpopulation of high responders. It is noted that a high seroresponse rate (>81.4%) was observed in participants with baseline nAb titres < LLOQ from both cohorts (V1222/B1222 and VmRNA/B1222), an observation that demonstrates the adequate boosting ability of AZD1222 dose, and that the lower seroresponse rate in the overall population was due to the difficulty to achieve seroresponse of ≥ 4-fold rise in nAb from baseline in subjects with higher pre-booster titres. Moreover, the AZD1222 booster effect in the overall population was demonstrated by a seroresponse rate (>71%) when the seroresponse rates was determined using as criterion a ≥2 fold rise in binding antibodies from baseline.

The MAH acknowledges that it has not followed the CHMP advice to compare the immune response of the VmRNA/B1222 group to an mRNA primary series treatment group. The MAH indicates that it was not possible to access serum samples for the VmRNA cohort nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study. Taking into account this explanation and the observations made in the previous paragraph, together the fact that the comparison made by the MAH in terms of GMT ratio showed higher titers in the population that received the AZD1222 booster as compared with a population in which clinical efficacy was shown (primary series of AZD1222), it is considered that the data submitted support the use of AZD1222 as a heterologous booster.

It is noted that very recently a preprint publication (not yet peer-reviewed) (https://www.medrxiv.org/content/10.1101/2022.04.29.22274483v1) (Effectiveness of ChAdOx1-S COVID-19 Booster Vaccination against the Omicron and Delta variants in England) from UK, provides evidence of protection against Omicron variant following homologous AZD1222 booster.

In conclusion, all the data provided by the MAH in response to the RSI provide clear support for the use of AZD1222 as a homologous or heterologous booster.

5. Clinical Safety aspects

5.1. Methods – analysis of data submitted

The main purpose of this submission is to provide the interim booster results from AZD1222 booster treatment in previously vaccinated cohorts (AZD1222 cohort and mRNA cohort) from Study D7220C00001. The interim analysis was conducted using a date of cut-off (DOC) of 11 October 2021.

In addition, the study D7220C00001 includes other multiple "cohorts" and treatment groups (AZD2816 booster in previously vaccinated with AZD1222 or mRNA vaccine; and COVID-19 vaccine naïve) that were not the focus of this submission. The safety results for the AZD2816 booster were not extensively discussed, however a brief summary of the results from AZD2816 was provided.

The safety analysis of AZD1222 booster dose in previously vaccinated cohorts (AZD1222 cohort and mRNA cohort) were conducted in Seronegative Safety Analysis Set. Overall, the results for the Safety Analysis Set, which also included seropositive participants, were consistent with those for the Seronegative Safety Analysis Set.

Safety was assessed by the evaluation of solicited adverse events (AEs) that are commonly associated with vaccinations (i.e., reactogenicity), unsolicited AEs, SAEs (including deaths), medically attended adverse events (MAAEs) and adverse events of special interest (AESIs); evaluation of biochemistry and haematology clinical laboratory tests, and vital signs.

Solicited AEs were self-reported via a diary card in all participants for 7 days following each vaccination. Unsolicited AEs were recorded for 28 days after each dose of AZD1222 booster.

Deaths, SAEs, MAAEs, AESIs, and COVID-19 AEs occurring post-Day 29 through DCO were also included in the analysis dataset.

5.2. Results

5.2.1. Patient exposure

The seronegative safety analysis set included a total of 1296 participants, of which 646 were randomised to the AZD1222 booster treatment group (347 in AZD1222 cohort and 299 in mRNA cohort). The Safety Analysis Set, included a small proportion of seropositive participants, of which 43 were randomised to the AZD1222 booster treatment group (20 in AZD1222 cohort and 23 in mRNA cohort).

All participants in mRNA cohort who received AZD1222 booster dose had been previously vaccinated with the BNT162b2 vaccine.

Demographic and Baseline Characteristics

Summary tables (Table 31 and Table 32) comparing demographic and baseline characteristics of the two groups had been reported by the MAH during procedure (see section 7. *).*

Table 31 Demographic characteristics (Seronegative Safety Analysis Set)

Characteristic/Statistic	V1222/ B1222 N = 347	VmRNA/ B1222 N = 299
Age (years), n [a]		
Mean	59.7	55.3
SD	13.64	13.24
Median	62.0	55.0
Min	30	30
Age group (years), n (%) [a]		
≥ 18 to < 65	186 (53.6)	220 (73.6)
≥ 65	161 (46.4)	79 (26.4)
Sex, n (%)		
Male	187 (53.9)	113 (37.8)
Female	160 (46.1)	186 (62.2)
Race, n (%)		
White	303 (87.3)	268 (89.6)
Black or African American	2 (0.6)	3 (1.0)
Asian	9 (2.6)	8 (2.7)
Mixed	0	2 (0.7)
Unknown	33 (9.5)	18 (6.0)
Ethnic group, n (%)		
Hispanic or Latino	6 (1.7)	3 (1.0)
Not Hispanic or Latino	300 (86.5)	271 (90.6)
Missing	41 (11.8)	25 (8.4)
Country, n (%)		
Poland	12 (3.5)	1 (0.3)
United Kingdom	335 (96.5)	298 (99.7)

[a] Age at randomisation.

Table 32 Baseline characteristics (Seronegative Safety Analysis Set)

Characteristic/Statistic	V1222/ B1222 N = 347	VmRNA/ B1222 N = 299
Time since previous vaccination (days), n [a]		
n	347	299
Mean	230.5	127.1
SD	92.88	28.66
Median	266.0	120.0
Min	63	47
Max	379	211
Primary vaccination dosing interval (days), n		
n	347	299
Mean	54.2	63.3
SD	20.33	18.63
Median	59.0	70.0
Min	25	21
Max	91	86
BMI (kg/m^2), n		
n	346	298
Mean	27.2	27.9
SD	5.19	5.86
Median	26.8	26.9
Comorbidity at baseline		
At least one [b]	162 (46.7)	141 (47.2)
BMI ≥ 30 kg/m^2	85 (24.5)	90 (30.1)
Significant CVD	87 (25.1)	63 (21.1)
Chronic lung disease	30 (8.6)	27 (9.0)
Diabetes	21 (6.1)	9 (3.0)
None	185 (53.3)	158 (52.8)

The majority of the participants were White (>87%) and were from the UK (>96%).

In the seronegative safety analysis set, AZD1222 booster treatment group of the previously vaccinated cohort, 53.6% and 73.6% participants were between 18-64 years of age, and 53.9% and 37.8% of participants were males in the AZD1222 cohort and mRNA cohort, respectively.

An imbalance regarding the age group was observed between the groups receiving the booster AZ1222. Most of participants previously vaccinated with mRNA were 18-64 years of age (73,6% vs 26% subjects >65 years). Nevertheless, 53% and 46% primovaccinated with AZ1222 were between 18-64y and >65y respectively.

In the group of primovaccinated with mRNA, a higher percentage of females than males were included in the study (62.3% and 37.8% respectively). Approximately, 46.7% of the participants in AZD1222

cohort and 47.2% in mRNA cohort had at least one comorbidity associated with an increased risk for COVID-19 at baseline, with 24.5% and 30.1% reporting a BMI ≥ 30 kg/m2, and 25.1% and 21.1% reporting a significant, cardiovascular disease, respectively.

In addition, a higher interval between AZD1222 booster dose and AZD1222 previous vaccination (median: 266 days) than AZD1222 booster dose and mRNA previous vaccination (median:120 days).

Duration of Follow-up

At the time of the DCO, all participants in the AZD1222 booster treatment group had completed their Day 29 visit. The median duration of follow-up was similar across both cohorts: 90.0 days and 88.0 days in AZD1222 and mRNA cohort, respectively. See Table 33 for more detail about duration of follow up.

Table 33 Duration of Follow-up (Seronegative Safety Analysis Set)

	Number of Participants				
Statistic	**V1222/ B1222 N = 347**	**V1222/ B2816 N = 349**	**VmRNA/ B1222 N = 299**	**VmRNA/ B2816 N = 301**	**Total N = 1296**
Duration (days)					
n	347	349	299	301	1296
Mean	85.7	85.5	87.5	87.3	86.4
SD	18.05	18.53	11.77	11.76	15.62
Median	90.0	91.0	88.0	88.0	89.0
Min	35	23 [a]	60	60	23
Max	107	107	104	104	107

[a] Two participants withdrew consent on Day 23 (see Listing 16.2.1.1).

Follow-up duration is calculated as the number of days from the study intervention to withdrawal from study, study completion, or the data cutoff date, whichever is earlier.

B1222, Participants receiving a third dose booster of AZD1222; B2816, Participant receiving a third dose booster of AZD2816; Max, Maximum; Min, Minimum; n, Number of participants in analysis; N, Number of participants per treatment group; SD, Standard deviation; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222

Source: Table 14.3.1.2

5.2.2. Solicited Adverse Events

In Seronegative Safety Analysis Set, any Solicited AEs within the first 7 days following after AZD1222 booster dose were reported by 78.1% and 89.9% of participants of AZD1222 or mRNA cohort, respectively. Most of the solicited AEs following AZD1222 booster dose were mild to moderate in severity. The incidence of Grade 3 solicited AE was 1.5% in AZD1222 cohort and 12.1% in mRNA cohort. No Grade 4 solicited AEs were reported.

Solicited AEs were reported at the highest frequency on Day 2 (the day after booster vaccination) with > 60% of participants reporting AEs in each cohort. Reported AEs steadily declined thereafter, with < 13% of participants in each cohort reporting a solicited AE at Day 8.

Table 34 Overall summary of solicited adverse events- through Day 8 (Seronegative Safety Analysis Set)

	Number of participants (%)	
	V1222/B1222 N = 347	VmRNA/B1222 N = 299
Any solicited AE	264 (78.1)	268 (89.9)
Grade 1	156 (46.2)	102 (34.2)
Grade 2	103 (30.5)	130 (43.6)
Grade 3	5 (1.5)	36 (12.1)
Grade 4	0	0
Any local solicited AE	208 (61.5)	227 (76.2)
Grade 1	163 (48.2)	144 (48.3)
Grade 2	44 (13.0)	79 (26.5)
Grade 3	1 (0.3)	4 (1.3)
Grade 4	0	0
Any systemic solicited AE	204 (60.4)	237 (79.5)
Grade 1	117 (34.6)	93 (31.2)
Grade 2	83 (24.6)	111 (37.2)
Grade 3	4 (1.2)	33 (11.1)
Grade 4	0	0
Any solicited AE, by study day		
Day 1	107 (39.1)	84 (32.7)
Day 2	215 (66.6)	245 (84.8)
Day 3	153 (48.1)	197 (69.1)
Day 4	105 (33.4)	148 (53.4)
Day 5	66 (21.0)	106 (39.4)
Day 6	49 (15.8)	77 (28.2)
Day 7	39 (12.5)	41 (15.1)
Day 8	23 (8.9)	29 (12.7)

Solicited local AEs

In the Seronegative Safety Analysis Set, solicited local AEs after AZD1222 booster dose were reported by 61.5% and 76.2% of participants of AZD1222 or mRNA cohort.

The most frequently reported solicited local injection site AEs within 7 days after AZD1222 booster dose were tenderness (54.4% in AZD1222 cohort vs 71.1% in mRNA cohort) and pain (37.9% vs 49.7%); following of swelling (3.6% vs 4.0%) and redness (3% vs 4.4%).

Most of the local AEs were mild or moderate in intensity. A 0.3% of subjects in AZD1222 cohort and 1.3% in mRNA cohort experienced grade 3 local AEs after AZD1222 booster dose. No grade 4 severe local AEs were reported in any participants receiving AZD1222 booster dose.

Solicited systemic AEs:

In the Seronegative Safety Analysis Set, solicited systemic AEs after AZD1222 booster dose were reported by 60.45% and 79.5% of participants of AZD1222 or mRNA cohort.

The most frequently reported solicited systemic injection site AEs within 7 days after AZD1222 booster dose were fatigue (42.0% in AZD1222 cohort vs 56.7% in mRNA cohort) and headache (33.7% vs 51.3%); other frequently reported systemic solicited AEs were muscle pain (32.1% vs 47.3%), malaise

(21.3% vs 41.6%), nausea (12.1% vs 22.1%) chills (5.0% vs 29.5%) and vomiting (0.6% vs 1.3%). Fever: (≥37.9ºC) was reported in 1.5% participants of AZD1222 cohort and in 10.1% participants of mRNA cohort.

Most of the local AEs were mild or moderate in intensity. A 1.2% of subjects in AZD1222 cohort and 11.1% in mRNA cohort experienced grade 3 systemic AEs after AZD1222 booster dose. No grade 4 severe systemic AEs were reported in any participants receiving AZD1222 booster dose. In mRNA cohort participants chills (5%), fatigue (5.4%) and headache (3.4%) were the grade 3 solicited systemic AEs mostly reported.

Table 35 Solicited Adverse Events by severity (Seronegative Safety Analysis Set)

	Number of participants (%)			
Grade: Severity	**V1222/B1222 n/N = 338/347**	**V1222/B2816 n/N = 344/349**	**VmRNA/B1222 n/N = 298/299**	**VmRNA/B2816 n/N = 298/301**
Local solicited AEs at the site of the injection				
Pain				
Any severity	128 (37.9)	150 (43.6)	148 (49.7)	169 (56.7)
1: Mild	121 (35.8)	138 (40.1)	131 (44.0)	135 (45.3)
2: Moderate	7 (2.1)	12 (3.5)	17 (5.7)	34 (11.4)
3: Severe	0	0	0	0
4: ER or hospitalisation	0	0	0	0
Redness				
Any severity	10 (3.0)	8 (2.3)	13 (4.4)	12 (4.0)
1: Mild (2.5-5 cm)	5 (1.5)	4 (1.2)	5 (1.7)	7 (2.3)

	Number of participants (%)			
Grade: Severity	**V1222/B1222 n/N = 338/347**	**V1222/B2816 n/N = 344/349**	**VmRNA/B1222 n/N = 298/299**	**VmRNA/B2816 n/N = 298/301**
2: Moderate (5.1-10 cm)	4 (1.2)	4 (1.2)	7 (2.3)	3 (1.0)
3: Severe (> 10 cm)	1 (0.3)	0	1 (0.3)	2 (0.7)
4: Necrosis or exfoliative dermatitis	0	0	0	0
Tenderness				
Any severity	184 (54.4)	196 (57.0)	212 (71.1)	220 (73.8)
1: Mild	143 (42.3)	151 (43.9)	139 (46.6)	137 (46.0)
2: Moderate	41 (12.1)	42 (12.2)	70 (23.5)	79 (26.5)
3: Severe	0	3 (0.9)	3 (1.0)	4 (1.3)
4: ER or hospitalisation	0	0	0	0
Swelling				
Any severity	12 (3.6)	11 (3.2)	12 (4.0)	12 (4.0)
1: Mild (2.5-5 cm)	8 (2.4)	8 (2.3)	5 (1.7)	7 (2.3)
2: Moderate (5.1-10 cm)	3 (0.9)	3 (0.9)	7 (2.3)	3 (1.0)
3: Severe (> 10 cm)	1 (0.3)	0	0	2 (0.7)
4: Necrosis	0	0	0	0
Systemic solicited AEs				
Fever				
Any severity	5 (1.5)	11 (3.2)	30 (10.1)	39 (13.1)
1: Mild (37.9-38.4 °C)	5 (1.5)	5 (1.5)	25 (8.4)	25 (8.4)
2: Moderate (38.5-38.9 °C)	0	5 (1.5)	4 (1.3)	9 (3.0)
3: Severe (39.0-40 °C)	0	1 (0.3)	1 (0.3)	5 (1.7)
4: > 40 °C	0	0	0	0
Chills				
Any severity	17 (5.0)	25 (7.3)	88 (29.5)	124 (41.6)
1: Mild	10 (3.0)	16 (4.7)	28 (9.4)	48 (16.1)
2: Moderate	7 (2.1)	8 (2.3)	45 (15.1)	55 (18.5)
3: Severe	0	1 (0.3)	15 (5.0)	21 (7.0)
4: ER or hospitalisation	0	0	0	0
Muscle pain				
Any severity	78 (23.1)	86 (25.0)	141 (47.3)	164 (55.0)
1: Mild	48 (14.2)	60 (17.4)	60 (20.1)	65 (21.8)

	Number of participants (%)			
Grade: Severity	**V1222/B1222 n/N = 338/347**	**V1222/B2816 n/N = 344/349**	**VmRNA/B1222 n/N = 298/299**	**VmRNA/B2816 n/N = 298/301**
2: Moderate	30 (8.9)	25 (7.3)	73 (24.5)	81 (27.2)
3: Severe	0	1 (0.3)	8 (2.7)	18 (6.0)
4: ER or hospitalisation	0	0	0	0
Fatigue				
Any severity	142 (42.0)	124 (36.0)	169 (56.7)	191 (64.1)
1: Mild	81 (24.0)	64 (18.6)	64 (21.5)	45 (15.1)
2: Moderate	58 (17.2)	52 (15.1)	89 (29.9)	116 (38.9)
3: Severe	3 (0.9)	8 (2.3)	16 (5.4)	29 (9.7)
4: ER or hospitalisation	0	0	0	1 (0.3)
Headache				
Any severity	114 (33.7)	114 (33.1)	153 (51.3)	170 (57.0)
1: Mild	89 (26.3)	89 (25.9)	83 (27.9)	87 (29.2)
2: Moderate	24 (7.1)	24 (7.0)	60 (20.1)	74 (24.8)
3: Severe	1 (0.3)	1 (0.3)	10 (3.4)	9 (3.0)
4: ER or hospitalisation	0	0	0	0
Malaise				
Any severity	72 (21.3)	93 (27.0)	124 (41.6)	147 (49.3)
1: Mild	47 (13.9)	63 (18.3)	57 (19.1)	58 (19.5)
2: Moderate	25 (7.4)	28 (8.1)	59 (19.8)	75 (25.2)
3: Severe	0	2 (0.6)	8 (2.7)	14 (4.7)
4: ER or hospitalisation	0	0	0	0
Nausea				
Any severity	41 (12.1)	31 (9.0)	66 (22.1)	62 (20.8)
1: Mild	34 (10.1)	27 (7.8)	46 (15.4)	44 (14.8)
2: Moderate	7 (2.1)	4 (1.2)	20 (6.7)	18 (6.0)
3: Severe	0	0	0	0
4: ER or hospitalisation	0	0	0	0
Vomiting				
Any severity	2 (0.6)	3 (0.9)	4 (1.3)	4 (1.3)
1: Mild	1 (0.3)	1 (0.3)	1 (0.3)	0
2: Moderate	1 (0.3)	2 (0.6)	3 (1.0)	4 (1.3)
3: Severe	0	0	0	0

	Number of participants (%)			
Grade: Severity	V1222/B1222 n/N = 338/347	V1222/B2816 n/N = 344/349	VmRNA/B1222 n/N = 298/299	VmRNA/B2816 n/N = 298/301
4: ER or hospitalisation	0	0	0	0

Percentages are based on n, the number of participants per category.

Through Day 8: This table includes solicited AEs with a protocol-defined collection period of 7 days post booster dose.

Participants with multiple occurrences in the same category are counted once per category regardless of the number of occurrences.

AE, Adverse event; B1222, Participants receiving a third dose booster of AZD1222; B2816, Participant receiving a third dose booster of AZD2816; ER, Emergency Room/Emergency Department/Accident & Emergency; n, Number of participants in analysis; N, Number of participants per treatment group; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222

Derived from: Table 14.3.2.2.2

5.2.3. Unsolicited Adverse events

In Seronegative Safety Analysis Set, any Unsolicited AEs within the first 28 days following after AZD1222 booster dose were reported by 22.5% and 23.7% of participants of AZD1222 or mRNA cohort, respectively. Most of the unsolicited AEs following AZD1222 booster dose were mild to moderate in severity. No SAEs were reported within 28 days following after AZD1222.

Related AEs, as determined by the investigator, were reported by 5.2% and 7.7% in AZD1222 cohort and mRNA cohort. Most of them were mild-moderate in severity. The 0.3% of participants of each group reported Grade 3 related unsolicited AEs.

There were no clinically meaningful imbalances by SOC between the two groups. The unsolicited AEs by SOC were summarized in Table 36.

Table 36 Unsolicited adverse events by System Organ Class- through Day 29 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	V1222/B1222 N = 347	V1222/B2816 N = 349	VmRNA/B1222 N = 299	VmRNA/B2816 N = 301
Any unsolicited AE	78 (22.5)	66 (18.9)	70 (23.4)	75 (24.9)
Infections and infestations	22 (6.3)	12 (3.4)	10 (3.3)	15 (5.0)
Nervous system disorders	13 (3.7)	8 (2.3)	12 (4.0)	24 (8.0)
General disorders and administration site	10 (2.9)	12 (3.4)	17 (5.7)	15 (5.0)

	Number of participants (%)			
	V1222/B1222 N = 347	V1222/B2816 N = 349	VmRNA/B1222 N = 299	VmRNA/B2816 N = 301
Musculoskeletal and connective tissue	14 (4.0)	13 (3.7)	13 (4.3)	13 (4.3)
Gastrointestinal disorders	11 (3.2)	11 (3.2)	13 (4.3)	7 (2.3)
Respiratory, thoracic and mediastinal	9 (2.6)	4 (1.1)	7 (2.3)	5 (1.7)
Injury, poisoning and procedural complications	7 (2.0)	7 (2.0)	6 (2.0)	8 (2.7)
Skin and subcutaneous tissue disorders	8 (2.3)	6 (1.7)	3 (1.0)	3 (1.0)
Investigations	5 (1.4)	5 (1.4)	3 (1.0)	5 (1.7)
Eye disorders	2 (0.6)	3 (0.9)	2 (0.7)	2 (0.7)
Blood and lymphatic system disorders	1 (0.3)	2 (0.6)	4 (1.3)	2 (0.7)
Vascular disorders	0	2 (0.6)	4 (1.3)	3 (1.0)
Reproductive system and breast disorders	3 (0.9)	1 (0.3)	1 (0.3)	3 (1.0)
Metabolism and nutrition disorders	2 (0.6)	3 (0.9)	0	1 (0.3)
Ear and labyrinth disorders	1 (0.3)	2 (0.6)	1 (0.3)	1 (0.3)
Renal and urinary disorders	2 (0.6)	0	0	1 (0.3)
Immune system disorders	2 (0.6)	0	0	0
Cardiac disorders	0	2 (0.6)	0	0
Hepatobiliary disorders	1 (0.3)	0	0	0
Psychiatric disorders	0	0	0	1 (0.3)

By preferred term (PT), the most frequently reported AEs (i.e., Headache and Fatigue) were commonly reported following vaccine administration

Table 37 Most common (≥ 10 events) Unsolicited Adverse events by Preferred term – Through day 29 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	V1222/B1222 N = 347	V1222/B2816 N = 349	VmRNA/B1222 N = 299	VmRNA/B2816 N = 301
Any unsolicited AE	78 (22.5)	66 (18.9)	70 (23.4)	75 (24.9)
Headache	4 (1.2)	4 (1.1)	7 (2.3)	13 (4.3)
Fatigue	4 (1.2)	9 (2.6)	7 (2.3)	5 (1.7)
Diarrhoea	1 (0.3)	2 (0.6)	8 (2.7)	3 (1.0)
Myalgia	4 (1.2)	4 (1.1)	5 (1.7)	1 (0.3)
Oropharyngeal pain	5 (1.4)	3 (0.9)	4 (1.3)	0
Arthralgia	2 (0.6)	4 (1.1)	2 (0.7)	4 (1.3)

Regarding the most common unsolicited AEs considered as related to AZD1222 booster vaccine, fatigue, headache, myalgia, and vaccination site lymphadenopathy were commonly reported as side-effects of vaccinations in general and known adverse drug reactions for AZD1222. There were 2 related AEs of Fibrin D-dimer increased in mRNA cohort (0.7%) comparing to zero in AZD1222 cohort (the total events of Fibrin D-dimer increased were reported in participants randomised to a single study site).

Table 38 Most common (≥ 4 events and at least 1 event in each group) related unsolicited adverse events by Preferred term – Through day 29 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	V1222/B1222 N = 347	V1222/B2816 N = 349	VmRNA/B1222 N = 299	VmRNA/B2816 N = 301
Any related unsolicited AE	18 (5.2)	13 (3.7)	23 (7.7)	24 (8.0)
Fatigue	1 (0.3)	2 (0.6)	4 (1.3)	3 (1.0)
Headache	1 (0.3)	0	3 (1.0)	3 (1.0)
Fibrin D dimer increased	0	1 (0.3)	2 (0.7)	3 (1.0)
Myalgia	2 (0.6)	2 (0.6)	2 (0.7)	0
Vaccination site lymphadenopathy	0	0	3 (1.0)	2 (0.7)
Thrombocytopenia	0	1 (0.3)	3 (1.0)	0

5.2.4. SAEs and Deaths

5.2.4.1. Deaths

There were no deaths reported after AZD1222 booster dose through data cut-off.

5.2.4.2. Other serious Adverse Events

There were no SAEs through Day 29 reported after AZD1222 booster dose.

There were 4 SAEs reported by 3 AZD1222-vaccinated participants after AZD1222 booster dose through data cut-off. All four were reported in the AZD1222 cohort and none of them were reported during the first 4 weeks following administration.

The SAEs included Small intestinal obstruction, Herpes zoster oticus, Diabetic neuropathy, and Hypoglossal nerve paralysis. Of these SAEs, Diabetic neuropathy, and Hypoglossal nerve paralysis were reported by the same participant, whose hypoglossal nerve paralysis was also an AESI (see 5.2.5.1).

None of the reported SAEs were assessed as related to study intervention by the investigator.

5.2.5. Adverse Events of Special Interest

Predefined AESIs were neurologic events, vascular events, hematologic events, and potential immune-mediated conditions.

In the Seronegative Safety Analysis Set, there were 6 AESIs reported through Day 29, 5 of them in the mRNA vaccinated cohort, and 3 additional AESIs reported through data cut-off.

Table 39 Adverse events of Special interest by Category and Preferred Term (Seronegative Safety Analysis Set)

	Number of Participants (%)	
	V1222/B1222 N = 347	VmRNA/B1222 N = 299
Through Day 29		
Any unsolicited AESI	1 (0.3)	5 (1.7)
Haematologic	0	3 (1.0)
Thrombocytopenia	0	3 (1.0)
Neurologic	1 (0.3)	1 (0.3)
Paraesthesia	1 (0.3)	1 (0.3)
PIMC – Musculoskeletal disorders	0	1 (0.3)
Arthritis reactive	0	1 (0.3)
Through Data Cutoff		
Any unsolicited AESI	2 (0.6)	7 (2.3)
Haematologic	0	3 (1.0)
Thrombocytopenia	0	3 (1.0)
Neurologic	1 (0.3)	2 (0.7)
Paraesthesia	1 (0.3)	2 (0.7)
PIMC – Musculoskeletal disorders	0	2 (0.7)
Arthritis reactive	0	2 (0.7)
PIMC – Neuroinflammatory disorders	1 (0.3)	0
Hypoglossal nerve paralysis	1 (0.3)	0

	Number of Participants (%)	
	V1222/B1222 N = 347	VmRNA/B1222 N = 299
Through Day 29		
Any unsolicited AESI	1 (0.3)	5 (1.7)
Haematologic	0	3 (1.0)
- Thrombocytopenia	0	3 (1.0)
Neurologic	1 (0.3)	
- Paraesthesia	1 (0.3)	1 (0.3)
PIMC – Musculoskeletal disorders	0	1 (0.3)
- Arthritis reactive	0	1 (0.3)
Through Data Cutoff		
Any unsolicited AESI	2 (0.6)	7 (2.3)
Haematologic	0	3 (1.0)
- Thrombocytopenia	0	3 (1.0)
Neurologic	1 (0.3)	2 (0.7)
- Paraesthesia	1 (0.3)	2 (0.7)
PIMC – Musculoskeletal disorders	0	2 (0.7)
- Arthritis reactive	0	2 (0.7)
PIMC – Neuroinflammatory disorders	1 (0.3)	0
- Hypoglossal nerve paralysis	1 (0.3)	0

None of these AESIs met the criteria for referral to the Neurological AESI Expert Committee or Haematology Expert Panel.

There was one additional AESI of Sensory disturbance in a seropositive participant of AZD1222 cohort (related, non-serious, mild intensity, started on Day 5).

During the procedure (see section 7) the MAH provided the AESI´s narratives and a summary indicating whether they were considered related to the vaccine or the treatment. In Safety analysis Set (including seronegative and seropositive participants) there were 2 related AESIS in AZD1222 cohort (paraesthesia and sensory disturbance events) and 4 related AESIS in mRNA cohort (3 thrombocytopenia events and 1 event of arthritis reactive).

None of related AESIS were serious. Brief descriptions are as follows:

Paraesthesia

Intermittent mild paraesthesia that started on Day 1 in the hand in one participant of AZD1222 cohort following administration of the AZD1222 booster in the deltoid. The event is ongoing. Updated product information for Vaxzevria including paraesthesia (and hypoaesthesia) as an adverse drug reaction was submitted on 7 Mar 2022 with the latest PBRER procedure.

Sensory disturbance

Mild sensory disturbance (hot and cold feeling) in knee and foot was reported by one seropositive participant of AZD1222 cohort after AZD1222 booster dose. The event was reported to be intermittent and resolved spontaneously. The event was judged to be related to study treatment.

Thrombocytopenia

There were 3 non-serious, related thrombocytopenia AEs in the mRNA cohort. None of the participants had an associated thrombotic or bleeding event. All events were transient and resolved without treatment. Intensity was reported as mild for 1 of the AEs and moderate for the other 2 AEs. Dates of onset were 9, 10 and 29 days from date of booster dose. The AE on Day 9 was reported as worsening of known thrombocytopenia. In no participant were platelets recorded as falling below 100 x 10^9/L.

Arthritis reactive

One participant of mRNA cohort reported a non-serious AESI of new onset multiple joint pain. The event was considered as related to study intervention. Arthralgia is listing as an adverse drug reaction in the Vaxzevria prescribing information.

Only 1 AESI was considered serious but not related. It was reported by a participant from AZD1222 cohort with an ongoing medical history of diabetes. The participant reported on Day 46 an event of paresthesia, arthralgia, myalgia and elevated glycated hemoglobin (AE: Diabetic neuropathy, grade 2, not related). On Day 48, the participant reported worsening symptoms and was diagnosed with neuropathy secondary to hyperglycaemia (severity grade 3; SAE: Diabetic neuropathy). On Day 54 the participant was diagnosed with a neurological disorder (severity grade 3; SAE: neurological disorder), which was considered secondary to poorly controlled diabetes.

In addition, there were 7 AEs of COVID-19 after AZD1222 booster dose, 4 cases in AZD1222 cohort and 3 in mRNA cohort. All COVID-19 cases were non-serious, mild or moderate, and none were related to study treatment. These cases provide no evidence that AZD1222 is associated with enhanced respiratory disease.

5.2.6. MAAEs

Medically attended AEs were 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 for any reason.

Within 28 days post AZD1222 booster dose, 8.4% and 5.0% of participants from AZD1222 and mRNA cohort reported any MAAE; 2.3% and 2.4% of participants reported an additional MAAE through the data cut-off.

The most common MAAEs by PT were Urinary tract infection (n=5), Hypertension (n=4), Headache (n=3), and Ear infection (n=3).

No notable findings for MAAEs and no clinically meaningful imbalances in the incidence of MAAEs by SOC or PT across AZD1222 and mRNA cohort were reported.

The frequencies of MAAEs considered related to the vaccine, reported by the MAH, were very low and similar between cohorts (1.1% and 0.9% in AZD1222 and mRNA cohort, respectively).

No imbalance was observed, and the narrative of the events did not raise any additional safety concern.

5.2.7. Laboratory findings

The MAH had reported during procedure tables summarizing laboratory and haematological parameters (Table 40 and Table 41).

Table 40 Summary of Coagulation and Haematology results over time (Seronegative Safety Analysis Set)

Parameter Treatment group	N	Baseline	Day 29	Change from baseline	
		Mean	Mean	Mean change	SD
Activated Partial Thromboplastin Time (sec)					
V1222:B1222	347	26.701	26.646	0.014	1.6197
VmRNA:B1222	299	26.902	26.642	-0.183	1.2877
Basophils (10^9/L)					
V1222:B1222	347	0.043	0.044	0.001	0.0240
VmRNA:B1222	299	0.043	0.041	-0.002	0.0199
D-Dimer (mg/L)					
V1222:B1222	347	0.3475	0.4455	0.0010	0.7902
VmRNA:B1222	299	0.3077	0.3305	0.0178	0.4268
Eosinophils (10^9/L)					
V1222:B1222	347	0.1603	0.1742	0.0138	0.1167
VmRNA:B1222	299	0.1603	0.1713	0.0106	0.0726
Fibrinogen (umol/L)					
V1222:B1222	347	9.3652	9.2925	-0.0540	1.4657
VmRNA:B1222	299	9.1566	9.3111	0.1611	1.3977

Table 41 Summary of Coagulation and Haematology results over time (Seronegative Safety Analysis Set)

Parameter Treatment group	N	Baseline	Day 29	Change from baseline	
		Mean	Mean	Mean change	SD
Hemoglobin (g/L)					
V1222:B1222	347	141.39	140.48	-1.18	5.928
VmRNA:B1222	299	139.38	138.98	-0.57	5.208
Leukocytes (10^9/L)					
V1222:B1222	347	6.301	5.928	-0.372	1.2794
VmRNA:B1222	299	6.416	6.203	-0.214	1.1577
Lymphocytes (10^9/L)					
V1222:B1222	347	1.796	1.782	-0.006	0.3238
VmRNA:B1222	299	1.837	1.812	-0.020	0.3414
Monocytes (10^9/L)					
V1222:B1222	347	0.5201	0.4930	-0.0277	0.1279
VmRNA:B1222	299	0.5053	0.4940	-0.0109	0.1103
Neutrophils (10^9/L)					
V1222:B1222	347	3.783	3.424	-0.373	1.0592
VmRNA:B1222	299	3.865	3.678	-0.194	1.0079
Platelets (10^9/L)					
V1222:B1222	347	255.18	251.33	-3.53	25.757
VmRNA:B1222	299	257.21	255.97	-1.10	27.468
Prothrombin Time (sec)					
V1222:B1222	347	10.752	10.715	-0.024	0.4802
VmRNA:B1222	299	11.142	10.991	-0.133	0.4544

Table 42 Summary of chemistry results over time (Seronegative Safety analysis set)

Parameter Treatment group	N	Baseline	Day 29	Change from baseline	
		Mean	Mean	Mean change	SD
Alanine Aminotransferase (IU/L)					
V1222:B1222	347	23.60	23.13	-0.57	13.210
VmRNA:B1222	299	22.85	22.95	0.21	6.771
Alkaline Phosphatase (IU/L)					
V1222:B1222	347	70.10	70.48	0.48	9.935
VmRNA:B1222	299	73.48	74.22	0.45	8.344
Aspartate Aminotransferase (IU/L)					
V1222:B1222	347	24.50	24.74	0.19	11.643
VmRNA:B1222	299	23.63	23.92	0.05	6.072
Bilirubin (umol/L)					
V1222:B1222	347	9.9590	10.2080	0.2509	3.6758
VmRNA:B1222	299	9.5779	9.4912	-0.1077	2.9583
Creatinine (umol/L)					
V1222:B1222	347	75.5978	75.0584	-0.6499	7.6282
VmRNA:B1222	299	72.8311	72.3772	-0.3517	7.6377

There were no clinically relevant mean changes in haematology or clotting parameters over time, including for platelets or D-dimer. There were no clinically relevant mean changes in clinical chemistry values over time, including for ALT, AST, ALP, bilirubin, or creatinine.

5.2.8. Safety in special populations

The study excluded individuals with immunosuppressive or immunodeficient disorders including HIV, history of thrombocytopenia and/or thrombosis, and neuroimmunology conditions.

There were no pregnancies during the study through the interim analysis database lock

5.2.9. Safety in Subgroups

The safety parameters were reviewed by subgroup for age at randomization, gender and comorbidity at baseline. In addition, during the procedure the MAH submitted the tables below with solicited AEs and unsolicited AEs by age group, sex, and comorbidity for study D7220C00001 for the seronegative safety analysis set (see section 7.).

5.2.9.1. Solicited AEs by subgroups

Age

In the AZD1222 booster treatment groups the safety profile was generally similar in both cohorts in older adults compared with younger adults 18 to 64 years of age, reporting the older group a reduced reactogenicity, as observed in previous clinical studies with AZD1222. Nevertheless, an imbalance regarding age was observed in demographic characteristics. A 46.4% and 26.4% were ≥65 year-aged in the AZD122 and mRNA cohort respectively. A total of 186 and 220 participants were between 18-64

years of age, and 161 and 79 were ≥65 years of age (AZD122 and mRNA cohorts respectively) (Table 31 - demography characteristics).

Table 43 Overall summary of solicited adverse events by age group for treatment groups receiving an AZD1222 Booster- through day 8 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	Age 18 to 64 years		Age 65 years and older	
	V1222/B1222 N = 186	VmRNA/B1222 N = 220	V1222/B1222 N = 161	VmRNA/B1222 N = 79
Any solicited AE	160 (87.4)	210 (95.5)	104 (67.1)	58 (74.4)
Grade 1	85 (46.4)	68 (30.9)	71 (45.8)	34 (43.6)
Grade 2	70 (38.3)	112 (50.9)	33 (21.3)	18 (23.1)
Grade 3	5 (2.7)	30 (13.6)	0	6 (7.7)
Grade 4	0	0	0	0
Any local solicited AE	138 (75.4)	191 (86.8)	70 (45.2)	36 (46.2)
Grade 1	100 (54.6)	119 (54.1)	63 (40.6)	25 (32.1)
Grade 2	37 (20.2)	69 (31.4)	7 (4.5)	10 (12.8)
Grade 3	1 (0.5)	3 (1.4)	0	1 (1.3)
Grade 4	0	0	0	0
Any systemic solicited AE	128 (69.9)	187 (85.0)	76 (49.0)	50 (64.1)
Grade 1	71 (38.8)	64 (29.1)	46 (29.7)	29 (37.2)
Grade 2	53 (29.0)	95 (43.2)	30 (19.4)	16 (20.5)
Grade 3	4 (2.2)	28 (12.7)	0	5 (6.4)
Grade 4	0	0	0	0
Any solicited AE, by study day				
Day 1	81 (53.3)	71 (36.8)	26 (21.3)	13 (20.3)
Day 2	137 (78.7)	195 (92.0)	78 (52.3)	50 (64.9)
Day 3	99 (58.6)	162 (77.9)	54 (36.2)	35 (45.5)
Day 4	68 (41.5)	122 (60.7)	37 (24.7)	26 (34.2)
Day 5	43 (25.6)	85 (43.1)	23 (15.6)	21 (29.2)
Day 6	24 (14.5)	61 (31.0)	25 (17.2)	16 (21.1)
Day 7	23 (14.1)	33 (16.7)	16 (10.7)	8 (11.0)
Day 8	16 (11.9)	24 (14.7)	7 (5.6)	5 (7.7)

Comorbidity

In the seronegative safety analysis set, 46.8% of the participants had at least one comorbidity associated with an increased risk for COVID-19 at baseline (obesity, i.e., BMI ≥ 30 kg/m2 at baseline, significant cardiovascular disease, chronic lung disease and diabetes). The reactogenicity of an AZD1222 booster dose was similar in participants independently of having or not comorbidities at baseline.

Table 44 Overall Summary of Solicited Adverse Events by Comorbidity for Treatment Groups Receiving an AZD1222 Booster – Through Day 8 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	At least one comorbidity		No comorbidity	
	V1222/B1222 N = 162	VmRNA/B1222 N = 141	V1222/B1222 N = 185	VmRNA/B1222 N = 158
Any solicited AE	117 (74.5)	119 (84.4)	147 (81.2)	149 (94.9)
Grade 1	61 (38.9)	45 (31.9)	95 (52.5)	57 (36.3)
Grade 2	51 (32.5)	57 (40.4)	52 (28.7)	73 (46.5)
Grade 3	5 (3.2)	17 (12.1)	0	19 (12.1)
Grade 4	0	0	0	0
Any local solicited AE	84 (53.5)	91 (64.5)	124 (68.5)	136 (86.6)
Grade 1	62 (39.5)	60 (42.6)	101 (55.8)	84 (53.5)
Grade 2	21 (13.4)	29 (20.6)	23 (12.7)	50 (31.8)
Grade 3	1 (0.6)	2 (1.4)	0	2 (1.3)
Grade 4	0	0	0	0
Any systemic solicited AE	96 (61.1)	107 (75.9)	108 (59.7)	130 (82.8)
Grade 1	49 (31.2)	40 (28.4)	68 (37.6)	53 (33.8)
Grade 2	43 (27.4)	52 (36.9)	40 (22.1)	59 (37.6)
Grade 3	4 (2.5)	15 (10.6)	0	18 (11.5)
Grade 4	0	0	0	0
Any solicited AE, by study day				
Day 1	46 (36.5)	37 (31.1)	61 (41.2)	47 (34.1)
Day 2	87 (58.4)	110 (80.3)	128 (73.6)	135 (88.8)
Day 3	70 (47.3)	81 (60.9)	83 (48.8)	116 (76.3)
Day 4	50 (33.3)	57 (42.9)	55 (33.5)	91 (63.2)
Day 5	30 (20.0)	41 (31.1)	36 (21.8)	65 (47.4)
Day 6	23 (16.0)	26 (19.7)	26 (15.7)	51 (36.2)
Day 7	23 (15.9)	15 (11.8)	16 (9.6)	26 (18.1)
Day 8	13 (10.3)	18 (16.7)	10 (7.6)	11 (9.2)

Sex

In the AZD1222 booster treatment groups, the overall incidence of solicited AEs reported within 7 days after a booster dose of AZD1222 was lower and less severe in males than in females across both cohorts. Nevertheless, a higher percentage of females were included in the mRNA group (62.2% vs 37.8% males) (Table 31- demography characteristics).

Table 45 Overall Summary of Solicited Adverse Events by Sex for Treatment Groups Receiving an AZD1222 Booster – Through Day 8 (Seronegative Safety Analysis Set)

	Number of participants (%)			
	Male		**Female**	
	V1222/B1222 N = 187	**VmRNA/B1222 N = 113**	**V1222/B1222 N = 160**	**VmRNA/B1222 N = 186**
Any solicited AE	129 (70.5)	93 (83.0)	135 (87.1)	175 (94.1)
Grade 1	88 (48.1)	40 (35.7)	68 (43.9)	62 (33.3)
Grade 2	39 (21.3)	44 (39.3)	64 (41.3)	86 (46.2)
Grade 3	2 (1.1)	9 (8.0)	3 (1.9)	27 (14.5)
Grade 4	0	0	0	0
Any local solicited AE	94 (51.4)	77 (68.8)	114 (73.5)	150 (80.6)
Grade 1	86 (47.0)	60 (53.6)	77 (49.7)	84 (45.2)
Grade 2	8 (4.4)	16 (14.3)	36 (23.2)	63 (33.9)
Grade 3	0	1 (0.9)	1 (0.6)	3 (1.6)
Grade 4	0	0	0	0
Any systemic solicited AE	99 (54.1)	79 (70.5)	105 (67.7)	158 (84.9)
Grade 1	62 (33.9)	33 (29.5)	55 (35.5)	60 (32.3)
Grade 2	35 (19.1)	38 (33.9)	48 (31.0)	73 (39.2)
Grade 3	2 (1.1)	8 (7.1)	2 (1.3)	25 (13.4)
Grade 4	0	0	0	0
Any solicited AE, by study day				
Day 1	46 (31.1)	18 (18.4)	61 (48.4)	66 (41.5)
Day 2	95 (54.6)	84 (77.1)	120 (80.5)	161 (89.4)
Day 3	72 (41.9)	63 (58.9)	81 (55.5)	134 (75.3)
Day 4	49 (28.3)	45 (44.1)	56 (39.7)	103 (58.9)
Day 5	30 (16.9)	33 (32.7)	36 (26.1)	73 (43.5)
Day 6	22 (13.2)	22 (20.8)	27 (18.9)	55 (32.9)
Day 7	21 (12.4)	12 (11.8)	18 (12.6)	29 (17.2)
Day 8	12 (8.3)	8 (9.1)	11 (9.7)	21 (15.0)

5.2.9.2. Unsolicited AEs by subgroups

In Table 46 unsolicited AEs by subgroup are summarized. A low percentage of AESIs or unsolicited AEs ≥ grade 3 severe AEs were observed. Therefore, it was difficult to make any conclusion regarding the unsolicited AEs assessed by subgroups.

Table 46 Unsolicited Adverse Events by Subgroup – Through Day 29 (Seronegative Safety Analysis Set)

	Number of participants/Number in analysis set (%)	
	V1222/B1222	**VmRNA/B1222**
Any unsolicited AE	78/347 (22.5)	70/299 (23.4)
By age group		
18-64 years	44/186 (23.7)	57/220 (25.9)
≥ 65 years	34/161 (21.1)	13/79 (16.5)
By sex		
Male	36/187 (19.3)	26/113 (23.0)
Female	42/160 (26.3)	44/186 (23.7)
By comorbidity		
At least one	45/162 (27.8)	35/141 (24.8)
None	33/183 (17.8)	35/158 (22.2)

5.2.10. Immunological events

Immunologic events were considered AESIs. Very few immunological disorders were observed. Three events were reported in the V1222/B1222 (dust allergy and seasonal allergy). None of them were related to the booster vaccine. No immunological events were reported in the other groups. No anaphylactic shock or hypersensitivity disorders were reported either.

5.2.11. Safety related to drug-drug interaction

The safety, immunogenicity, and efficacy of co-administration of AZD1222 with other vaccines have not been evaluated.

5.2.12. Discontinuation due to AEs

There were no discontinuation or withdrawal in participants who received AZD1222 booster vaccine in AZD1222 cohort or in mRNA cohort.

5.2.13. Post marketing experience

Some countries recently began administering booster doses of AZD1222 following a primary 2-dose series of AZD1222 in select populations. These include the UK, Chile, Dominican Republic, El Salvador, Guatemala, Honduras, Malaysia, Philippines, and Uruguay. The sporadic reporting of AEs following third dose boosters has not identified any new safety concerns.

5.3. Discussion

The main purpose of this submission is to provide the interim results of AZD1222 booster dose in previously vaccinated cohorts (AZD1222 cohort and mRNA cohort) from Study D7220C00001. The interim analysis was conducted using a date of cut-off (DOC) of 11 October 2021. In addition, the study included other cohorts with AZD2816 vaccine (a modified AZD1222 vaccine targeted against the Beta variant of SARS-CoV-2) in previously vaccinated subjects (with AZD1222 or mRNA vaccines). However, the MAH is not looking for the licensure for AZD2816, therefore, the MAH only included a brief summary of the results from AZD2816 booster groups.

In study D7220C00001 there were 1379 participants, of which, 689 received AZD1222 booster vaccine. From 689 participants, 367 were previously vaccinated with AZD1222 and 322 with mRNA vaccine. All participants previously vaccinated with mRNA vaccine had received the BNT162b2 vaccine.

Of the total of 689 participants, 43 were seropositive (20 in AZD1222 cohort and 23 in mRNA cohort).

The safety analysis submitted by the MAH was conducted in Seronegative Safety Analysis Set who received AZD1222 booster vaccine (N=646). From theses, 646 participants, 347 participants were previously vaccinated with AZD1222 and 299 participants were previously vaccinated with a mRNA vaccine. The median number of follow-up days after AZD1222 booster dose was similar in both groups up to the cut-off date (90 days in AZD1222 cohort and 88 days in mRNA cohort). Of note, there were no discontinuation or withdrawal in participants of both cohorts.

Demographic and Baseline Characteristics

The MAH has submitted during the procedure summary tables comparing baseline demographic characteristics of the two groups (V1222/B1222 and VmRNA/B1222) of the Seronegative safety analysis set.

The majority of the participants were white and were from the UK. An imbalance regarding age and gender was observed in demographic and baseline characteristics in participant receiving AZD1222 booster dose. In the cohort previously vaccinated with AZD122, 53.6% were between 18-64 years of age vs 73.6% in the mRNA cohort. Of note is that only a 26% of subjects >65y had been primo vaccinated with mRNA. Moreover, a higher percentage of females were included in the mRNA group (62.2% females vs 37.8% males) compared AZD1222 group (46.1% vs 53.9%, respectively).

In addition, there was a larger interval between AZD1222 booster dose and AZD1222 previous vaccination (median: 266 days) than AZD1222 booster dose and mRNA previous vaccination (median:120 days). No differences were observed in the percentage of participants with comorbidity (46.7% and 47.2% in AZD1222 and mRNA cohort, respectively).

The imbalance observed regarding age, gender and the dose interval between the booster dose and previous vaccination may mask the observed differences between the two groups regarding reactogenicity. As a response to the RSI submitted during the procedure, the MAH concludes that the differences in demographic and baseline characteristics between two cohorts may explain some, but not all, of the differences in reactogenicity between them, and suggests that with more balanced cohorts of previously vaccinated AZD1222 or mRNA participants, there may have been a smaller difference in the reactogenicity profile of the AZD1222 cohort versus the mRNA cohort.

Solicited Adverse Events

In the Seronegative Safety Analysis Set, the profile of solicited local and systemic AEs was similar in both groups and not different to the known local and systemic reactogenicity described for AZD1222, although the frequency of each solicited local and systemic AEs was higher in mRNA cohort than in AZD1222 cohort (89.9% and 78.1% respectively). Most of the solicited AEs following AZD1222 booster

dose in both cohorts were mild to moderate in severity. However, the incidence of Grade 3 solicited AEs was much higher in mRNA cohort (12.1%) than in AZD1222 cohort (1.5%), mainly due to the higher incidence of solicited grade 3 systemic AEs observed in the mRNA cohort (11.1% vs 1.2%). As a response to the RSI, the MAH suggests that the severity of solicited AEs after AZD1222 booster dose in subjects previously vaccinated with mRNA was similar to that in naïve participants receiving a primary vaccination with AZD1222. However, taking into account the pooled data submitted during the MAA, the incidence of severe solicited systemic AEs was 6.6% after 1st dose and 2.2% after 2nd dose of AZD1222. Therefore, the severity of solicited systemic AEs after AZD1222 vaccination seems to be somewhat higher in people previously vaccinated with mRNA compared to people naïve who received the 1st dose of AZD1222

No significant differences in severity in solicited local AEs were observed between the two cohort groups (0.3% and 1.3% in AZD1222 and mRNA cohort, respectively).

It is known, from the data reported in the pooled University of Oxford studies and the D8110C00001 study, that the incidence of solicited AEs is lower after the second dose than after a first dose of primary vaccination with AZD1222. However, with the data reported in this submission it is not possible to define whether the safety pattern of the homologous booster increases or decreases with respect to the first or second dose of AZD1222.

Moreover, the data reported in this submission indicate that heterologous booster with AZD1222 in subjects previously vaccinated with an mRNA vaccine elicits a higher reactogenicity than the homologous booster with AZD1222.

The MAH has provided, as requested in RSI (see section 9.), a comparative table of the incidence of reactogenicity events following homologous and heterologous AZD1222 booster versus primary AZD1222 vaccination. According to the response submitted by MAH, the proportion of participants receiving a heterologous booster with a solicited AE was generally similar than the proportion of participants receiving the primary dose series of AZD1222.

Additionally, according to the data from the Pooled Oxford Studies (submitted during conditional MA) the incidence of solicited AEs is higher after the first dose than after the second dose of AZD1222 during the primary series.

With the analysis now provided by the MAH, it is not possible to determine whether the reactogenicity after homologous or heterologous AZD1222 booster is similar or not to the known reactogenicity profile (regarding frequencies) after 1st or 2nd dose of AZD1222.

Unsolicited Adverse events

In the Seronegative Safety Analysis Set, the frequency of unsolicited AEs following AZD1222 booster dose was similar in both cohorts (22.5% and 23.7% in AZD1222 and mRNA cohorts, respectively). The majority of them were not considered as related to AZD1222 booster dose by the investigators. However, the frequency of Related Unsolicited AEs following AZD1222 booster was higher in mRNA cohort (7.7%) than in AZD1222 cohort (5.2%). Most of the unsolicited AEs following AZD1222 booster dose in both cohorts were mild to moderate in severity and no differences were observed in the incidence of Grade 3 related unsolicited AEs between cohort groups (0.3% each).

There were no clinically meaningful imbalances in unsolicited AEs by SOC between two groups. There was a slight imbalance in frequencies of SOC general disorders and administration site (2.9% vs 5.7%) and nervous system disorder (3.7% vs 4.0%) between AZD1222 and mRNA cohorts, respectively.

The most frequently related AEs reported (≥4 events) were fatigue, headache, myalgia, vaccination site lymphadenopathy, thrombocytopenia and increased Fibrin D-dimer. All these AEs, except elevated Fibrin D-dimer, are included in section 4.8 of the current SmPC.

The MAH has provided, as it has been requested in RSI, a discussion about whether people previously vaccinated with mRNA who would receive a booster dose of AZD1222, could have an increased risk of vaccination site lymphadenopathy. The MAH indicated that the subjects receiving a AZD1222 booster dose after mRNA primary vaccination may be at a somewhat higher risk of vaccination site lymphadenopathy compared to previously AZD1222 vaccinated individuals receiving their third AZD1222 dose. However, the incidence reported in the mRNA cohort is in line with that reported for individuals that received a primary AZD1222 vaccination as disclosed in the current SmPC.

In addition, two AEs of increased Fibrin D-dimer considered as related to AZD1222 booster dose were observed in the mRNA cohort (0.7%) compared to zero in the AZD1222 cohort. The MAH provided as a response to RSI a report of the two resolved events of increases in D-dimer in the mRNA cohort. The narratives provided do not raise any safety concern and the not inclusion of elevated fibrin D-dimer as adverse drug reaction in the SmPC is justified.

Deaths, SAEs and MAAEs

No deaths were reported. Four SAEs were reported after the AZD1222 booster dose through the data cut-off, one of these was an AESI. None of the reported SAEs were considered related to AZD1222 booster dose.

The incidence of MAAEs reported after the booster dose was low and the frequencies similar in both groups (10.7% and 7.4% in AZD1222 and mRNA cohort, respectively). The most common MAAEs by PT were Urinary tract infection, Hypertension, Headache, and Ear infection.

The MAH has submitted the incidences of MAAEs considered as related to the vaccine, as requested in RSI. The frequencies of MAAEs considered related to the vaccine were very low and similar between both cohort groups (1.1% and 0.9% in AZD1222 and mRNA cohort, respectively).

Adverse Events of Special Interest

There were 6 AESIs reported through Day 29, and 3 additional AESIs reported through data cut-off in the seronegative safety cohort.

Of the 7 AESIS in the mRNA cohort 3 were mild events of thrombocytopenia without associated thrombotic or bleeding events and 1 was an event of arthralgia (reactive arthritis), all 4 were considered related to the vaccination.

There were 2 related AESIs in the AZD1222 cohort one event of paraesthesia and one of mild sensory disturbance in a seropositive participant.

Paraesthesia, thrombocytopenia and arthralgia are already included in Vaxzervria SmPC as adverse drug reactions.

Only the AESIs of neuropathy (diabetic) and hypoglossal nerve paralysis reported from the AZD1222 cohort were considered as SAEs. These events started on day 46 and were considered unrelated to the vaccine and secondary to a previously diagnosed diabetes.

There were 4 cases of COVID-19 in the AZD1222 cohort and 3 in mRNA cohort after the booster that were considered AESIs, none of which had characteristics of enhanced disease.

No immunological events related to the booster dose of AZ1222 were reported.

MAAEs

The incidence of MAAEs reported after the booster dose was low and the frequencies similar in both groups (10.7% and 7.4% in AZD1222 and mRNA cohort, respectively). The most common MAAEs by PT were Urinary tract infection, Hypertension, Headache, and Ear infection.

The MAH has submitted the incidences of MAAEs considered as related to the vaccine during the procedure. The frequencies of MAAEs considered related to the vaccine were very low and similar between both cohort groups (1.1% and 0.9% in AZD1222 and mRNA cohort, respectively).

Laboratory findings

The MAH provided the laboratory and haematological parameters, as requested in RSI. The laboratory, haematological and chemistry shift were similar between both cohorts. The majority of laboratory, haematological and chemistry parameters were within normal clinical range and did not raise any safety concerns

Safety in Subgroups

In Seronegative Safety Analysis Set, the safety profile was assessed by age, gender and comorbidity at baseline. There was not an analysis regarding the serostatus at baseline. In addition, a brief description of the safety in the Safety Analysis Set (which included seronegative and seropositive participants) was reported by the MAH. Although, the data of seropositive participants were limited, the safety results of AZD1222 booster dose in all participants were consistent with those for the Seronegative Safety Analysis and no new safety signal was observed.

Regarding the reactogenicity profile of AZD1222 booster dose in the Seronegative Safety Analysis Set, higher frequencies of solicited local and systemic AEs were reported in the mRNA cohort than in the AZD1222 cohort, when assessed by age, comorbidity and sex groups. These data were similar than the data provided from the overall population.

Nevertheless, some differences in the reactogenicity profile were observed in the different subgroups:

- Solicited (local and systemic) and unsolicited AEs were milder and reported less frequently in adults aged ≥65 years compared with adults aged 18 to 64 years in each cohort groups. These results were consistent to data reported in previous AZD1222 studies. Nevertheless, it should be considered that an imbalance was found regarding the number of participants by age.
- The incidence and severity of solicited and unsolicited AE was higher in female than in male after AZD1222 booster dose in both cohorts. This profile was in line with the results observed in previous AZD1222 studies.
- The incidences of solicited (local and systemic) AEs and unsolicited AEs were slightly lower in participants with comorbidity at baseline than in those without comorbidity at baseline. However, this difference was not deemed clinically relevant and was in line with results observed in previous AZD1222 studies.

Post marketing experience

Some countries recently began administering booster doses of AZD1222 following a primary 2-dose series of AZD1222 in select populations. These include the UK, Chile, Dominican Republic, El Salvador, Guatemala, Honduras, Malaysia, Philippines, and Uruguay. The sporadic reporting of AEs following third dose boosters has not identified any new safety concerns.

6. Changes to the Product Information

As a result of this variation, sections 4.2, 4.4, 4.8 and 5.1 of the SmPC are being updated to introduce

a booster dose in individuals aged 18 years and older.

Section 4.2 is also updated to remove the statement about the interchangeability of Vaxzevria with other COVID-19 vaccines to complete the vaccination course.

The package leaflet (PL) is updated accordingly. Please refer to Attachment 1.

7. Assessment of the responses to the Rapporteur's preliminary list of questions requested during the procedure

In order to facilitate the rapid assessment of this variation, as requested by the MAH, a preliminary list of clinical questions (10 efficacy, 4 safety) were raised to the MAH on the 9th of March during preparation of the preliminary assessment report. By the 23rd of March, responses to 4 out of 10 efficacy questions and to all 4 safety questions were provided. In this situation, the MAH was informed that the eight preliminary questions answered by the MAH would be included and assessed in the first assessment report and that the remaining 6 question would be included as RSI (section 8.), as well as any other new questions. The responses provided by the MAH have been discussed in this assessment Report.

Efficacy

1) The MAH should indicate whether the pseudoneutralisation assay and the wild type virus neutralization assay used in trial D7220C00001 are the same described at the time of the initial MAA of Vaxzevria. Any differences should be justified.

Summary of MAH's Response:

The pseudoneutralisation assay and the wild type virus (live virus) neutralisation assay used in study D7220C00001 are the same assays as described in the initial MAA for Vaxzevria. The live virus neutralisation assay was validated prior to testing of study samples in study D7220C00001.

Assessment of MAH's response:

The MAH confirmed that the pseudoneutralisation assay and the wild type virus (live virus) neutralisation assay used in study D7220C00001 are the same assays as described in the initial MAA for Vaxzevria.

Point solved

2) The validation report of the PhenoSense Anti-SARS-COV-2 Pseudovirus Neutralising Antibody Assay (Wuhan-Hu-1) should be provided.

Summary of MAH's Response:

The validation report for PhenoSense Anti-SARS-COV-2 Pseudovirus Neutralising Antibody Assay (Wuhan-Hu-1) is provided as Appendix A and the bridging validation report for B.1.351 (Beta variant) is provided as Appendix B.

Assessment of MAH's response:

The validation report for Neutralising Antibody Assay (Wuhan-Hu-1) has been provided.

Conclusion: Point solved

3) Apparently the sample size calculations were made before the CHMP recommendation (made in the FAL) to prepare three independent SAPs. The MAH is asked to comment on whether sample size was updated once the three independent SAPs were prepared. The question focuses only on pre-vaccinated subjects (with either AZD1222 or an mRNA vaccine) that received a booster of AZD1222.

Summary of MAH's Response:

The sample size calculations were not updated for the individual sub-SAPs. At the time of the CHMP recommendation, the data were already locked and enrolment had completed. As such, AstraZeneca did not update the sample size after being unblinded to treatment arm. Given the updated testing hierarchies to be used at the primary analysis, the sample sizes within each sub-SAP provide adequate power for hypothesis testing.

Assessment of MAH's response:

The MAH acknowledged that the sample size was not updated since the data were already locked, and enrolment had completed. The Applicant has clarified that the sample sizes within each sub-SAP provide adequate power for hypothesis testing. This is considered acceptable, and the issue can be considered clarified.

Conclusion: Point solved

4) One important aspect that has not been followed by the MAH is the recommendation made by the CHMP in the second FAL in order to include a claim in the product information that AZD1222 can be used to boost the response in persons previously vaccinated with COVID-19 mRNA vaccines. The advice stated "*To demonstrate the adequacy of the immune response following the booster dose in this cohort, the company should plan a head-to-head comparison in which the response following a AZD1222 or AZD2816 booster is demonstrated to be non-inferior to the response with an mRNA booster. Alternatively, the response 28 days following the AZD1222 or AZD2816 booster response is demonstrated to be non-inferior to the response 28 days after the two primary doses of mRNA vaccine*." Instead of following the CHMP advice, the MAH compared the response following AZD1222 booster to the historical control group that received two doses of AZD1222. The MAH is asked to comment on the justification for not following the CHMP advice.

Summary of MAH's Response:

AstraZeneca acknowledge that data from a primary vaccination scheme with an mRNA vaccine or AZD1222 non-inferiority data to an mRNA booster are not available from study D7220C00001, nor from any other study performed by AstraZeneca. However, the University of Oxford-sponsored Com-COV study that evaluated primary series of AZD1222 and BNT162b2 in a homologous and heterologous schedule (Liu et al 2021) and the University of Southampton-sponsored COV-BOOST study (Munro et al 2021) provide data using the same validated assays that support the use of an AZD1222 booster dose following a primary vaccination scheme with an mRNA-based vaccine. While some baseline characteristics of participants, notably the average age, differed between the studies, Com-COV and COV-BOOST provide publicly available comparator data on heterologous boosting of AZD1222 following an mRNA-based primary vaccination scheme.

Com-COV was a randomised, controlled non-inferiority study conducted at 8 centres in the UK. Participants had no or well controlled comorbidities and had no history of laboratory-confirmed SARS-CoV-2 infection. The immunology cohort of participants (n = 100) were randomised 1:1:1:1 to receive primary vaccination series of AZD12222/AZD12222, AZD12222/BNT162b2 (Pfizer-BioNtech), BNT162b2/BNT162b2, or BNT162b2/AZD12222 with an interval of 28 days between the doses. The mean

age in participants receiving an AZD1222/AZD1222 primary series was 58.2 years (SD 4.81) and the mean age in participants receiving a BNT162b2/BNT162b2 primary series was 58.2 (SD 4.85).

COV-BOOST was a multicentre, randomised, controlled, Phase II study of third-dose booster vaccination against COVID-19. Participants had no history of laboratory-confirmed SARS-CoV-2 infection. Participants had received a primary vaccination series of AZD1222/AZD1222 or BNT162b2/BNT162b2. At least 70 days had passed after the second of 2 doses of AZD1222 or at least 84 days after the second of 2 doses of BNT162b2. Participants were generally older than those from the Com-COV study; the mean age in participants receiving an AZD1222/AZD1222 primary series was 63.7 (SD 14.1) and the mean age in participants receiving a BNT162b2/BNT162b2 primary series was 61.9 (SD 16.6).

For participants receiving a homologous AZD1222 booster dose, both spike binding as well as pseudoneutralising antibody responses were numerically higher after the AZD1222 booster in COV-BOOST than the titres observed in the primary series in Com-COV. For participants receiving a heterologous AZD1222 booster dose after a primary series of BNT162b2/BNT162b2 in COV-BOOST, SARS-CoV-2 anti-spike IgG titres were similar to those of participants who had received only a primary vaccination series with BNT162b2/BNT162b2 in Com-COV, while pseudotype neutralising antibody titres were higher for participants receiving a heterologous AZD1222 booster dose (Table 1).

Given that participants in the COV-BOOST study were older than those in the Com-COV study, it is likely that the COV-BOOST participants would have a diminished immune response compared with the Com-COV participants. It is therefore notable that SARS-CoV-2 anti-spike IgG and pseudotype neutralising antibody titres in participants who had received an AZD1222 booster still exceeded the corresponding titres in the younger participants receiving the BNT162b2/BNT162b2 primary series in Com-COV.

In the D7220C00001 study, immunobridging of spike-binding and neutralising antibody data in participants heterologously boosted with AZD1222 after a primary series of an mRNA vaccine to those vaccinated with a primary series of AZD1222 allows the comparison of neutralising antibody titres post-boost to a regimen that is shown to be clinically protective after a primary series. Hence, the matched historical controls from study D8110C00001 were appropriately used as a reference arm in analysis of data from the mRNA cohort.

While correlates of protection may be different between vaccine platforms, it is notable that heterologous AZD1222 boosters increased humoral responses and showed non-inferiority to a clinically effective vaccine regimen (AZD1222 primary series).

It is AstraZeneca's view that these data, in combination with immunogenicity data from D7220C00001, are sufficient to demonstrate the benefit of AZD1222 as a heterologous booster dose.

Table 1 **SARS-CoV-2 Anti-spike IgG and Pseudotype Neutralising Antibody Following a Primary Vaccination Series in the Com-COV Study and Following a Primary Vaccination Series and an AZD1222 Booster in the COV-BOOST Study**

		AZD1222 / AZD1222 (Com-COV)	AZD1222 / AZD1222 + AZD1222 (COV-BOOST)	BNT162b2 / BNT162b2 (Com-COV)	BNT162b2 / BNT162b2 + AZD1222 (COV-BOOST)
SARS-CoV-2 anti-spike IgG	n	105	99	110	97
	GMC (95% CI) in ELU/mL	1387 (1186-1623)	2457 (2058-2933)	13938 (12358-15719)	13424 (11702-15399)
Pseudotype neutralising antibody	n	101	98	102	98
	GMT (95% CI) in NT50	61 (50-73)	193 (161-231)	574 (475-694)	950 (802-1126)

CI, confidence interval; GMC, geometric mean concentration; GMT, geometric mean titre; ELU, ELISA laboratory unit; IgG, Immunoglobulin G; NT_{50}, 50% neutralising antibody titre
Derived from: Liu et al 2021 (the Com-COV study) and Munro et al 2021 (the COV-BOOST study)

Assessment of MAH's response:

The MAH's response followed the same original rationale and further discussed the data from the COV-BOOST study in support of deviation from the CHMP recommendation.

The MAH also compares the titres reached, 29 days after last vaccination, in participants from Com-COV and COV-BOOST, and they conclude that after receiving an homologous AZD1222 booster dose, both spike binding as well as pseudoneutralising antibody responses were numerically higher after the AZD1222 booster in COV-BOOST than the titres observed in the primary series in Com-COV (Table 1). It is however noted, that similar binding antibody titres were observed in subjects that received two doses of BNT162b2 as compared to those primed with two doses of BNT162b2 and later boosted with AZD1222. Thus, the interpretation of this comparison has to be taken with caution. Moreover, it is noted that a detailed comparison of baseline characteristics of the cohorts Com-COV and COV-BOOST are not discussed by the MAH, which adds another complication to interpret the results obtained from this comparison.

Conclusion: Issue not solved. Further pursued in RSI.

Safety

Question 5: Baseline demographics of AZD1222 booster groups Summary tables comparing baseline demographic characteristics of the two groups (V1222/B1222 and VmRNA/B1222) of Seronegative safety analysis set are missing and should be provided (similar to immunogenicity Tables 8 and 9).

Summary of MAH's Response:

The MAH provided table 2 and table 3 of demographic and baseline characteristics for the seronegative safety analysis set. Compared to the seronegative immunogenicity analysis set, results for which were presented in CSR Section 10.4, the seronegative safety analysis set only included an additional 25/1271 (1.9%) participants. There are no meaningful differences in participant characteristics between the seronegative immunogenicity analysis set and the seronegative safety analysis set

Assessment of MAH's response:

Tables of demographic and baseline characteristics for the Seronegative Safety Analysis Set were submitted by the MAH. The relevant information were included and discussed on section 8.2.1 and 8.3

Conclusion: Point solved

Question 6: Summary of adverse events of special interest: Summary of Annex 16.2.7.4 AESIs should be provided, including only AESIs reported in groups V1222/B1222 and VmRNA/B1222. In addition, it should be indicated whether they were considered related to the vaccine or the treatment. The narratives should be provided.

Summary of MAH's Response:

The MAH reported a summary and evaluation of AESIs in the V1222/B1222 and VmRNA/B1222 treatment groups, summary tables of Annex 16.2.7.4 by treatment group and narratives for the events.

Among 689 participants that received an AZD1222 booster, there were 17 AESIs reported. Of which, 7 AESIS (4 cases in the V1222/B1222 group and 3 cases in the VmRNA/B1222 group) were COVID-19. All COVID-19 cases were non-serious, mild or moderate, and none were related to study treatment. These cases provide no evidence that AZD1222 is associated with enhanced respiratory disease.

The other AESIs reported after AZD1222 booster dose were:

- 3 cases of paraesthesia, all non-serious and mild: 1 considered to be related to study treatment in the V1222/B1222 group, and 2 considered to be unrelated in the VmRNA/B1222 group (a participant reported tingling of arm along with shoulder pain on Day 49 and a participant described a sensation of pins and needles in shoulder on Day 20 attributable to poor ergonomics).
- 1 serious case in the V1222/B1222 group in a participant, who also reported diabetic neuropathy. The AESI was considered by the investigator to be secondary to poorly controlled diabetes and unrelated to treatment.
- 1 case of sensory disturbance in the AZD1222/B1222 group that was considered to be related to study treatment.
- 3 cases of thrombocytopenia in the VmRNA/B1222 group that were considered to be related study treatment.
- 2 cases of reactive arthritis reported in the VmRNA/B1222 group: 1 considered to be related to study treatment and 1 considered to be unrelated (attributed to a viral infection).

Assessment of MAH's response:

Summary tables of Annex 16.2.7.4 by treatment group including whether they were considered related to the vaccine or the treatment and narratives for the events were submitted by the MAH. The relevant information was included and discussed on section 8.2.5 and 8.3

Conclusion: Point solved.

Question 7: Safety subgroup data: Safety subgroup data should be summarized and provided

Summary of MAH's Response:

AstraZeneca provided summaries of solicited AEs and Unsolicited AEs by age group, sex, and comorbidity for study D7220C00001, for the seronegative safety analysis set.

Subgroup data are also available in CSR Section 14 for the full safety analysis set, which included seropositive participants. Only data from the treatment groups receiving a booster dose of AZD1222 are provided; the source tables in CSR Section 14 include data for the AZD2816 treatment groups.

Assessment of MAH's response:

Tables of solicited and unsolicited by age group, sex, and comorbidity for study D7220C00001 for the seronegative safety analysis set were provided by the MAH. The relevant information was included and discussed on section 8.2.9 and 8.3

Conclusion: Point solved

Question 8: Summary tables of safety parameters Tables summarizing laboratory and haematological parameters should be provided.

Summary of MAH's Response:

The MAH submitted a table summarizing results of coagulation and haematology parameters over time and another table with a summary of chemistry results over time in study D7220C00001 (the source tables in CSR Section 14 include data for the AZD2816 treatment groups).

Assessment of MAH's response:

The MAH provided tables summarizing laboratory and haematological parameters. However, the tables provided did not indicate the percentage and total number of participants with normal values, increased or decreased values of different laboratory and haematological parameters for groups V1222:B1222 and VmRNA:B1222 as requested by the assessor. Therefore, the MAH should provide again the tables with the indicated format and a discussion of the observed differences if they are

present.

Conclusion: Point not solved. Further pursued in RSI

8. Request for supplementary information (RSI)

8.1. Major objections

Clinical aspects

Efficacy

1) The data provided by the MAH are not considered sufficient to provide conclusive efficacy evidence to support de use of Vaxzevria as a booster dose, in adults 18 years and older, who were previously vaccinated with a primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based). The supplementary information that needs to be provided is the following:
    a) *A report showing adequate correlation between the pseudovirus neutralising and live virus neutralising assays is requested.* Immunogenicity comparisons were made based on pseudovirus neutralising antibodies. Thus, a report data showing good correlation of the pseudovirus neutralising and live virus neutralising assays should be provided. It is anticipated that this report would include data from a set of sera analysed by the two methods. In particular, the MAH should discuss the fact the seroresponse rate is very different for the two assays (96.8% vs 59.7%), and this result questions good concordance between both assays. Alternatively, results based on the wild type neutralizing assay could be submitted
    b) V*arious analyses to try explaining the failure to meet the key secondary endpoint (seroresponse)*. The Key secondary endpoint regarding the difference in seroresponse rates (when using the pseudoneutralization assay - Wuhan strain) was not met for any of the two groups (previously vaccinated with two doses of either mRNA or AZD1222 vaccine) that received an AZD1222 booster. Importantly, these differences are also shown when data from binding antibodies are analysed. In fact, after two doses of AZD1222, seroconversion rates were 98.8% (Table 14.2.7.1) whereas the seroconversion rates were only 68.2% for the V1222/B1222 group and 36.7% for the VmRNA/B1222 group (Table 19 from the interim CSR). The MAH should discuss on the possibility that only a few number of high-responders to the booster vaccine are driving compliance with the non-inferiority of the primary endpoint based on the ratio of GMTs (see also questions raised as OCs). In this regard, reverse distribution cumulative curves, before and after the booster dose should be provided. Moreover, taking into account that Wuhan baseline neutralising antibody titres (measured before administration of the booster) were below the LLoQ in approximately 75% of participants of the AZD1222 cohort and in 14% of participants in the mRNA cohort, the MAH is asked to determine the seroresponse rate in these subgroups in order to shed light on the booster effect of AZD1222.
    c) *Justification for not following the CHMP recommendation on the immunogenicity analysis to allow use of AZD122 as booster for those previously vaccinated with an mRNA vaccine.* The MAH did not follow the recommendation made by the CHMP on the comparisons to be made to support a claim for AZD1222 to be used to boost the response in persons previously vaccinated with COVID-19 mRNA vaccines. Instead, the MAH compared the response following AZD1222 booster to the historical control group that received two doses of AZD1222. The validity of this approach is questioned in that reaching the

primary endpoint is not a demanding requirement, considering that the baseline GMT of the VmRNA/B1222 group was 197 (95% CI 179-271)] which was quite close to the GMT reached after primary vaccination in subjects from the historical control group [242 (95% CI 224-262)]. Moreover, the fact that only 43% of the boosted subjects seroconverted, questions the claim that AZD1222 can be used to boost subjects that received a primary vaccination with an mRNA vaccine. The MAH is asked to comment.

8.2. Other concerns

Clinical aspects

Efficacy

2) In the document submitted to EMA by the MAH seeking advice on switching to use an historical control as the comparator arm for the previously vaccinated treatment groups it was stated: "*The inclusion/exclusion criteria of D7220C00001 and D8110C00001 are similar, and the data will not be confounded by dosing interval and the dose level, unlike the AZD1222 studies conducted by University of Oxford which included subjects who were vaccinated with 2 doses at various intervals (4 weeks to 16 weeks) and a subset of participants who were administered a low dose of AZD1222. […] Given that the dosing interval in the previously vaccinated group in the D7220C00001 study is 4 weeks, using data from studies with longer dosing interval is not appropriate in a trial designed to evaluate non-inferiority."*
In our understanding this text stated that the dosing interval (for the primary series of AZD1222) in the previously vaccinated group from trial D7220C00001 was going to be the same as in trial D8110C00001, which used an interval of 4 weeks. However, according to the data submitted by the MAH, the dosing interval in the previously AZD1222- vaccinated group in the study D7220C00001 varied from 25 to 91 days with a median of 59 days. The same situation applies to the previously vaccinated mRNA cohort (the interval between first and second dose ranged from 21 to 86 days with a median of 70 days). Moreover, as indicated above, the MAH stated that "using data from studies with longer dosing interval is not appropriate in a trial designed to evaluate non-inferiority", a sentence that questions the approach followed for the comparisons made in this variation application. The MAH is asked to comment on these statements.

3) Regarding the cohorts boosted with AZD1222, there is a wide time interval between first and second doses (see question 2), and also between second and booster dose (for the AZD 1222 cohort: 74 to 379 days with a median of 269 days; and for the mRNA cohort: 73 to 213 days with a median of 123 days). The MAH is asked to carry out a subgroup analysis to determine the impact of the dose interval in primary vaccination as well as the time interval from second to booster dose on the GMT and seroresponse rates reached.

4) The MAH states that: "Following a blinded review of protocol deviations related to the timing of doses, it was decided that only participants with intervals of < 70 days from second dose to booster dose … would be excluded from the immunogenicity analysis sets." Considering that usually for vaccines, booster doses are administered at 3-6 months after last vaccination, the MAH should provide the rationale for including participants with less than 90 days from second dose to booster dose in the analysis.

5) In total there were 13 seronegative participants who received the booster dose ≥ 70 days but < 90 days after their second dose, who were included in the Seronegative Immunogenicity Analysis

Set. The MAH is asked to clarify whether these 13 subjects received a booster dose of AZD1222 or AZD2816.

6) Based on the precision of the assay for detection of S-binding antibodies (Multiplexed ECL Method for the Detection of SARS-CoV-2 S) against the Wuhan strain, the MAH is asked to discuss the reliability of the data on seroconversion rate (based on a ≥2-fold increase) to assess the seroconversion rate achieved by the AZD1222 booster. If the MAH considers that the reliability is adequate, the MAH is asked to provide a Table with the percentage of seroresponders (≥2-fold) against the Wuhan strain in the cohorts boosted with AZD122 and calculate accordingly the serosresponse rate as compared to the historical control group.

7) In Tables 14 and 15 (interim CSR) it is indicated under superscript "a" that baseline titre for reference cohort: "For historical control, values at Day 29 after the first dose of the primary series (ie, prior to administration of the second dose of the primary series)". It is understood that this is a mistake and that the MAH is referring to values pre-dose 1. The MAH is asked to clarify.

8) In Table 15 from interim CSR, the baseline data for the Reference group (historical control group) are different from those sated in Table 14 for this same group. It is understood that the correct ones are those from Table 14. The MAH is asked to clarify.

9) The MAH should provide a comparative table of the patients previously vaccinated with two doses of AZD1222 from the study D8110C00001 (historical control group) and the patients recruited in this study (V1222/B1222 and VmRNA/B1222) with the standardised differences of the baseline characteristics for each groups of interest. The p-values should also be provided but only for exploratory purposes.

10) The MAH is asked to present a table comparing the differences in baseline and demographic characteristics between the subjects incorporated in the historical group from trial D8110C00001 and the subjects excluded from the historical group of the same trial. Additionally, another table with their standardised differences (including their p-values) should be provided.

11) The MAH provides a Table (Table 11) with few details regarding the historical control group. The MAH is asked to provide Tables with baseline and demographic characteristics (similar to Tables 8 and 9 from the Interim CSR) with three columns corresponding to the V1222/B1222, VmRNA/B1222 and the historical control groups in order to assess the good match of the AZD1222 boosted cohorts with the historical control group. In case differences are found between the historical control group and the two other cohorts (V1222/B1222 and VmRNA/B1222), the MAH is asked to discuss their impact in regard to the primary and key secondary immunogenicity analysis.

12) The MAH is asked to justify the proposed text in the SmPC that states "the third dose should be administered at least 3 months after completing the primary vaccination course" considering that the median time since second primary vaccination was 9 months in the AZD1222 cohort (range: 2.5 months to > 1 year), and 4 months in the mRNA cohort (range: 2.5 to 7 months).

Safety

13) Considering that there were differences in reactogenicity profile of AZD1222 booster dose between participants previously vaccinated with AZD1222 or mRNA vaccine by age and gender, the imbalance observed in demographic and baseline characteristics regarding age, gender and,

also, the dose interval (between the booster dose and previous vaccination) between the cohorts may mask the observed reactogenicity differences between the two groups in the overall population. The MAH should explain and justify if the imbalance in demographic and baseline characteristics observed between the two groups could contribute to the difference observed in the safety profile between two cohorts.

14) The incidence of Grade 3 solicited AEs was much higher in mRNA cohort (12.1%) than in AZD1222 cohort (1.5%), mainly due to the higher incidence of solicited grade 3 systemic AEs observed in the mRNA cohort (11.1% vs 1.2%). The severity of solicited systemic AEs after AZD1222 booster dose in mRNA cohort seemed to be higher than the primary vaccination series with AZD1222. The MAH should explain whether people previously vaccinated with mRNA who would receive a booster dose of AZD1222, could have an increased severity of reactogenicity comparing people naïve or previously vaccinated with AZD1222.

15) An assessment of the incidences of solicited AEs after the homologous or heterologous booster compared with AEs reported after primary AZD1222 vaccination should be provided.

16) The incidence of vaccination site lymphadenopathy in the mRNA cohort was higher (frequency as "common") than the incidence included in the current SmPC as "uncommon". The MAH should discuss whether people previously vaccinated with mRNA who would receive a booster dose of AZD1222, could have an increased risk of vaccination site lymphadenopathy.

17) Two AEs of increased Fibrin D-dimer considered as related to AZD1222 booster dose were observed in the mRNA cohort (0.7%) compared to zero in the AZD1222 cohort. The MAH should discuss these findings and justify whether or not an increase of elevated Fibrin D-dimer should be included in the SmPC for this population group.

18) There was no imbalance in the incidence of MAAEs after AZD1222 booster dose between AZD1222 and mRNA cohorts and no new safety signal was observed. However, the MAH has not reported on the evaluation of the relationship between the MAAEs and the investigational vaccine. This analysis should be submitted.
19) Regarding laboratory and haematological parameters, the MAH should provide the tables indicating the percentage and total number of participants with normal values, increased or decreased values of different laboratory and haematological parameters for groups V1222:B1222 and VmRNA:B1222.

20) Considering that trial D7220C00001 only recruited subjects > 30 yoa the MAH is asked to justify requesting a booster indication from 18 yoa. The risk of TTS in this population should also be taken into account.

9. Assessment of the responses to the request for supplementary information

9.1. Major objections

Clinical aspects

Efficacy

Question 1a: Correlation between pseudovirus and live virus neutralising assays

A report showing adequate correlation between the pseudovirus neutralising and live virus neutralising assays is requested. Immunogenicity comparisons were made based on pseudovirus neutralising antibodies. Thus, a report data showing good correlation of the pseudovirus neutralising and live virus neutralising assays should be provided. It is anticipated that this report would include data from a set of sera analysed by the two methods. In particular, the MAH should discuss the fact the seroresponse rate is very different for the two assays (96.8% vs 59.7%), and this result questions good concordance between both assays. Alternatively, results based on the wild type neutralizing assay could be submitted.

Summary of MAH's Response:

Results for a wild type neutralising assay based on samples from study D7220C00001 participants are now available and further demonstrate the correlation between the pseudoneutralisation and live virus neutralisation assays against Wuhan-Hu-1 strain. The results are reported below. But first, to fully address CHMP's concerns, the other 2 components of the request (for a report and for an explanation of the difference in assay results described in the CSP [96.8% vs. 59.7%]) are discussed.

Previously reported correlation analyses

Folegatti et al, 2020 reported on the concordance between nAbs as determined by the pseudovirus assay and live neutralisation assay for the Wuhan-Hu-1 strain. AstraZeneca subsequently assessed concordance for the Beta variant and reported the results in its 27 October 2021 Briefing Document.

Seroreponse rates quoted in the CSP

Regarding the seroresponse rates for both live and pseudovirus neutralisation assays quoted in the D7220C00001 Clinical Study Protocol (96.8% and 59.7%) these were obtained from historical data collected in the pooled Oxford studies (COV001, COV002, COV003, and COV005). Notably, there was no overlap in the subjects that contributed to these analyses. It is possible that with different assay summaries from separate subgroups, in addition to between subject variability and other prognostic factors known to cause differences in immunogenicity (such as age and baseline comorbitities), may make these reported seroresponse rates not comparable. Seroresponse rates from the pooled Oxford studies were included in Section 9.2 of the D7220C00001 CSP as estimates to support power calculations and were not intended as evidence of any correlation between assays.

New correlation analysis

AstraZeneca has now completed an analysis of concordance between the live virus neutralisation and pseudoneutralisation assays in a population of participants boosted with AZD1222. Data from the D7220C00001 study, summarised in Table 1, further demonstrate the correlation between the pseudoneutralisation and live virus neutralisation assays against Wuhan-Hu-1 strain. This new analysis was performed on live virus neutralisation assay data using serum samples from a subset of study

D7220C00001 that only became available after the interim database lock of 17 November 2021. The subset was chosen from the first 200 participants enrolled in each of the previously vaccinated with AZD1222 (ie, V1222) and previously vaccinated with an mRNA vaccine (ie, VmRNA) cohorts who were seronegative at baseline. The geometric mean fold rises among participants who had data for both assays were very similar in the V1222/B1222 and VmRNA/B1222 groups (live vs. neutralising assay GMFRs of 13.17 vs. 10.64 and 2.45 vs. 3.03 for the V1222 and VmRNA cohorts, respectively), as were the seroresponse rates (live vs. neutralising assay seroresponse of 83.5% vs. 81.3% and 31.0% vs. 39.3, respectively). This suggests that, within the same participants, concordance between these assays is quite high.

In addition, these two assays were determined to be well correlated when comparing quantitative results, with a Spearman rank correlation coefficient of 0.66 in both treatment groups. Spearman rank correlations represent the more conservative estimate of correlation but do not require a normality assumption regarding the distribution of the residuals. Overall, these data show very similar responses as the pseudovirus neutralisation assay and further support the use of the pseudovirus neutralisation assay for the assessment of nAb responses to AZD1222 vaccination.

Table 1 Correlation of live neutralisation and pseudoneutralisation assays against Wuhan-Hu-1 at Day 29 (Seronegative immunogenicity analysis set)

Statistic	V1222/B1222		VmRNA/B1222	
	Live neutralisation assay	Pseudoneutralisation assay	Live neutralisation assay	Pseudoneutralisation assay
n	92	92	89	89
GMT	1999.01	265.15	4816.72	1106.31
95% CI	(1666.65, 2397.64)	(212.86, 340.34)	(4171.92, 5561.18)	(915.16, 1337.39)
GMFR	13.17	10.64	2.45	3.03
95% CI	(10.41, 16.67)	(8.13, 13.94)	(1.98, 3.03)	(2.36, 3.89)
Seroresponse % (n/N)	83.5 (76 / 91)	81.3 (78 / 96)	31.0 (27 / 87)	39.3 (35 / 89)
95% CI	(74.3, 90.5)	(72.0, 88.5)	(21.5, 41.9)	(29.1, 50.3)
Correlation coefficient [a]	0.66		0.66	
95% CI	(0.53, 0.76)		(0.53, 0.76)	

[a] Spearman Rank correlation is used as the normal distribution assumption was rejected.
Titre values measured as below the lower limit of quantification (58 for live neutralisation, 40 for pseudoneutralisation) are imputed to a value that is half of the lower limit. Titre values measured as above upper limit of quantification (12,800 for live neutralisation, 787,339 for pseudoneutralisation) are imputed at the upper limit value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.
Fisher's z transformation is used to derive the confidence interval.
CI confidence interval; GMT geometric mean titre; n Number of subjects who have both live neutralisation and pseudoneutralisation titre data at Day 29.
Source: Appendix E, IEMT Tables 12.1.1 and 21.2.1.1

Assessment of MAH's response:

The MAH has clarified that the seroresponse rates for the live and pseudovirus neutralisation assays quoted in the D7220C00001 Clinical Study Protocol (96.8% and 59.7%, respectively) were not obtained using the same set of sera, and therefore they are not indicative of a lack of correlation of the two assays. This is agreed by the Assessors.

The MAH has now provided new data from an analysis of concordance between the live virus neutralisation and pseudoneutralisation assays for the Wuhan strain in a population of participants boosted with AZD1222 from trial D7220C00001. The data show good agreement of the two assays in terms of geometric mean fold rises and seroresponse rates. Moreover, when comparing quantitative results, the Spearman rank correlation coefficient was of 0.66 in both treatment groups. This figure, although not close to an optimal value of 0.9, it is considered to show an adequate correlation of the two assays with regard to the Wuhan strain.

In conclusion, the data provided show adequate correlation of the results obtained by the pseudovirus neutralisation assay and the live virus neutralisation assay. Thus, the use of the pseudovirus neutralisation assay in the trial D7220C00001 for the assessment of nAb responses after AZD1222 booster is considered justified.

Conclusion: Point solved

Question 1b: Seroresponse rate

Various analyses to try explaining the failure to meet the key secondary endpoint (seroresponse). The Key secondary endpoint regarding the difference in seroresponse rates (when using the pseudoneutralization assay - Wuhan strain) was not met for any of the two groups (previously vaccinated with two doses of either mRNA or AZD1222 vaccine) that received an AZD1222 booster. Importantly, these differences are also shown when data from binding antibodies are analysed. In fact, after two doses of AZD1222, seroconversion rates were 98.8% (Table 14.2.7.1) whereas the seroconversion rates were only 68.2% for the V1222/B1222 group and 36.7% for the VmRNA/B1222 group (Table 19 from the interim CSR). The MAH should discuss on the possibility that only a few number of high-responders to the booster vaccine are driving compliance with the non-inferiority of the primary endpoint based on the ratio of GMTs (see also questions raised as OCs). In this regard, reverse distribution cumulative curves, before and after the booster dose should be provided. Moreover, taking into account that Wuhan baseline neutralising antibody titres (measured before administration of the booster) were below the LLoQ in approximately 75% of participants of the AZD1222 cohort and in 14% of participants in the mRNA cohort, the MAH is asked to determine the seroresponse rate in these subgroups in order to shed light on the booster effect of AZD1222.

Summary of MAH's Response:

AstraZeneca considers that the antibody GMTs in study D7220C00001 post booster dose, despite not attaining a ≥ 4-fold increase, reached levels that provide increased protection. Further, an analysis excluding high responders has shown that these higher titres were not overly influenced by a small number of high-responding study participants.

Seroresponse

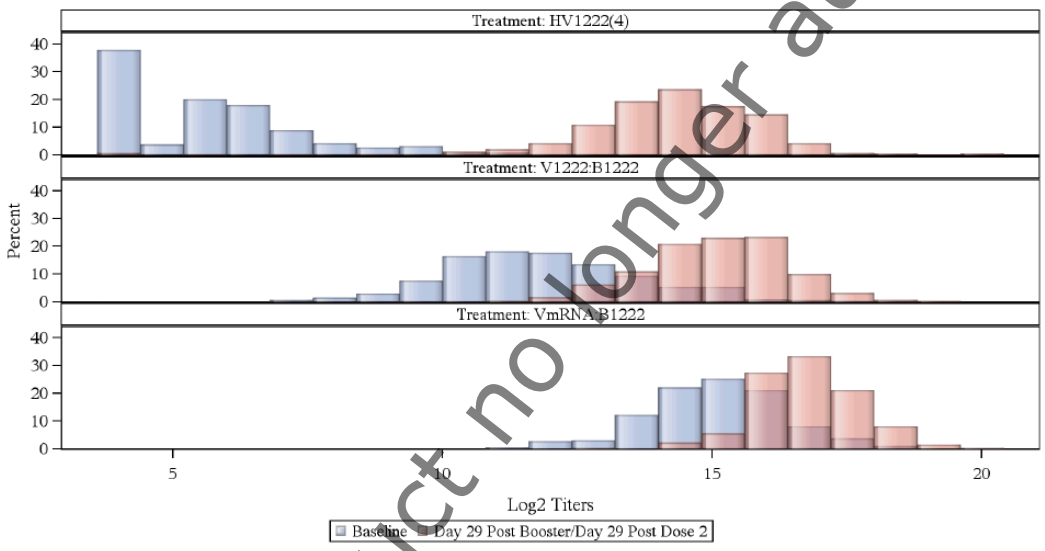
A seroresponse of at least 4-fold increase in antibody titres is considered a relevant outcome in a vaccine-naïve population in which baseline antibody levels are low or non-existent. AstraZeneca has postulated that in previously vaccinated individuals, with higher baseline antibodies, a < 4-fold rise in antibodies post booster dose may still be sufficient to reach a level considered to provide increased protection. AstraZeneca considers that the data from study D7220C00001 support this conclusion, with the immune responses induced by a booster dose of AZD1222 meeting or exceeding those elicited by the primary series against Wuhan-Hu-1 strain.

High responders

To address CHMP's concerns, the impact of high-responding study participants was investigated by multiple modalities, ie, histograms, applying Cook's distance, and reverse cumulative distribution curves.

As shown in Figure 1, the data for antibody titres follows a log-normal distribution of GMT at 28 days after baseline and do not show evidence of high responders skewing the distribution. If high-responders were driving the non-inferiority of GMT, a bimodal distribution would be expected, which is not evident in the data. Furthermore, this same figure illustrates that baseline titres are considerably lower in our historical control population, which demonstrates why a higher seroresponse is seen in this population. Moreover, these data demonstrate that participants with a previous mRNA vaccine have higher baseline titres, showing why a low seroresponse (36.7%) is seen in this population.

Figure 1 **Histograms of log_2 titres for Wuhan-Hu-1 spike protein binding for historical controls and participants boosted with AZD1222 (Seronegative Immunogenicity Analysis Set)**

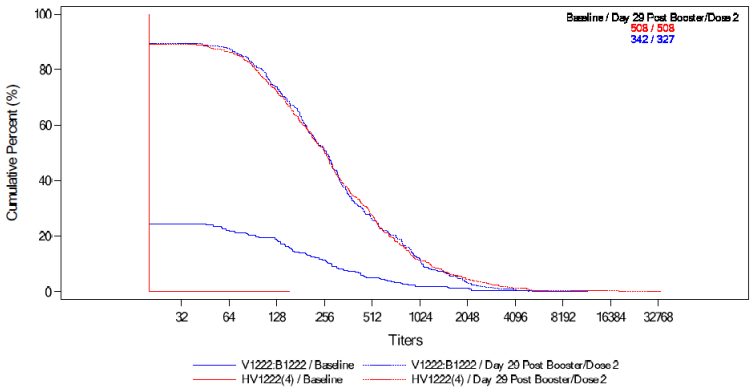


Titre values measured as below LLoQ (previously vaccinated = 23, historical controls = 33) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (previously vaccinated = 14,000,000, historical controls = 2,000,000) are imputed at the ULoQ value.
Historical control participants, ie, HV1222(4), were selected from those who had baseline and Day 29 post dose 2 pseudo neutralisation data.
B, booster vaccination; H, Historical Controls; LLoQ, Lower limit of quantification; ULoQ, Upper limit of quantification; V, primary vaccination
Source: Appendix F, IEMT Figure 13.1

A sensitivity analysis was performed that removed high responders exceeding a Cook's distance of > 4/n. The GMT ratio was marginally decreased in this analysis: to 0.96 (0.86, 1.07) from 1.03 (0.92, 1.15) for the V1222/B1222 group and to 2.97 (2.69, 3.29) from 3.08 (2.78, 3.41) for the VmRNA/B1222 group. However, non-inferiority would still have been met if these participants were removed and, therefore, the overall conclusions on immunogenicity remain unchanged.

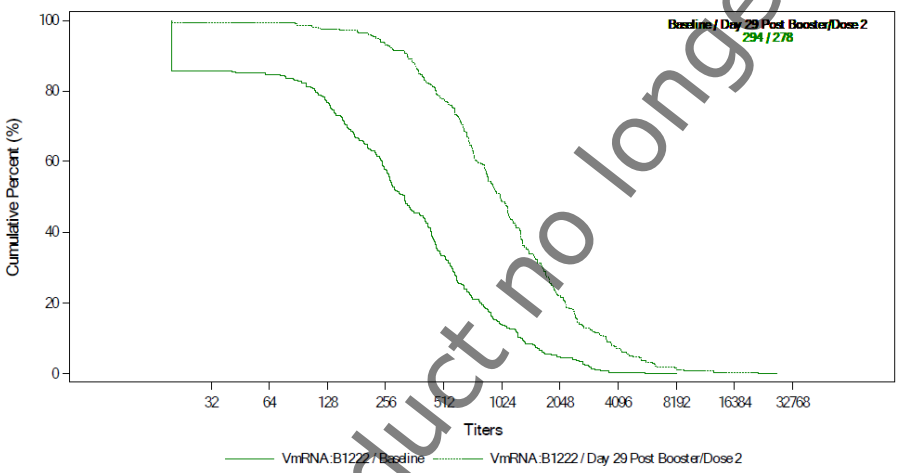
In addition, the requested reverse cumulative distribution curves (Figure 2 and 3) show that while approximately 75% of V1222 participants are < LLOQ at baseline, nearly all participants have increases in nAb titres post boost, and responses are log normal in distribution as well as having overlapping distribution with historic controls. Notably, for historic controls 100% of the V1222 participants had a baseline nAb titre < LLOQ. For VmRNA groups, a much lower proportion of participants are < LLOQ at baseline, but nAb titres are increased in nearly all participants post booster, with a notable shift in the curves seen post booster as compared to the baseline values.

Figure 2 **Reverse cumulative distribution plot of Wuhan-Hu-1 pseudoneutralising antibodies for participants previously vaccinated with AZD1222 and historical controls (Seronegative Immunogenicity Analysis Set)**



Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.
Historical control participants, i.e., HV1222(4), are selected from those who had baseline and Day 29 post dose 2 pseudo neutralisation data.
LLoQ Lower limit of quantification; ULoQ Upper limit of quantification; V primary vaccination; B booster vaccination; H Historical Controls.
Source: Appendix F, Figure 18.2.1.1

Figure 3 **Reverse cumulative distribution plot of Wuhan-Hu-1 pseudoneutralising antibodies for participants previously vaccinated with a mRNA vaccine (Seronegative Immunogenicity Analysis Set)**



Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.
LLoQ Lower limit of quantification; ULoQ Upper limit of quantification; V primary vaccination; B booster vaccination; H Historical Controls.
Source: Appendix F, Figure 18.2.2.1

Participants with antibodies < LLOQ

The seroresponse rates for pseudoneutralising antibodies after a booster dose of AZD1222 were evaluated in participants with baseline nAb titres < LLOQ. For both the V1222 and VmRNA groups, a higher rate of seroresponse was observed in participants with baseline nAb titres < LLOQ than in the overall population (Table 2). These data further emphasize that the seroresponse rate differences achieved after a booster dose are heavily influenced by the GMT at baseline. Importantly, while seroresponse rates were higher in those with baseline nAb titres < LLOQ, GMTs at Day 29 were higher in the overall population. This further demonstrates that high-responders were not driving non-inferiority for the primary endpoint. While those with baseline nAb titres < LLOQ were most likely to achieve seroresponse of ≥ 4-fold rise from baseline, those with higher baseline nAb titres also experienced a boost in GMTs to a level considered to provide increased protection.

Table 2 **Summary of Pseudoneutralising Antibody GMT and GMFR in Participants Previously Vaccinated with AZD1222 – Overall and Participants with Baseline nAb < LLOQ (Seronegative Immunogenicity Analysis Set)**

	Baseline	Day 15		Day 29	
Statistic		**GMT**	**Fold rise**	**GMT**	**Fold rise**
Overall Population: V1222/B1222 (Wuhan-Hu-1) N=342					
n	342	332	332	327	327
GMT	37.64	229.69	6.06	243.50	6.54
95% CI	33.05, 42.87	198.76, 265.44	5.17, 7.09	212.61, 278.88	5.60, 7.63
Seroresponse % (n^a/N^b)			61.4 (204 / 332)		66.1 (216 / 327)
Baseline nAb < LLOQ: V1222/B1222 (Wuhan-Hu-1) N=257					
n	257	248	248	247	247
GMT	20.00	185.84	9.29	203.12	10.16
95% CI	NE, NE	156.90, 220.14	7.84, 11.01	173.87, 237.29	8.69, 11.86
Seroresponse % (n^a/N^b)			76.2 (189 / 248)		81.4 (201 / 247)
Overall Population: VmRNA/B1222 (Wuhan-Hu-1) N=294					
n	294	274	274	278	278
GMT	269.71	862.42	3.28	1029.25	3.77
95% CI	230.56, 315.50	769.56, 966.49	2.84, 3.79	918.33, 1153.58	3.26, 4.37
Seroresponse % (n^a/N^b)			35.4 (97 / 274)		43.2 (120 / 278)
Baseline nAb < LLOQ: VmRNA/B1222 (Wuhan-Hu-1) N=41					
n	41	38	38	37	37
GMT	20.00	442.20	22.11	542.48	27.12
95% CI	NE, NE	279.14, 700.51	13.96, 35.03	374.06, 786.71	18.70, 39.34
Seroresponse % (n^a/N^b)			86.8 (33 / 38)		97.3 (36 / 37)

[a] n is the number of seroresponders.
[b] N is the number of subjects with a baseline measurement and an assessment at the given time point.
Seroresponse is defined as a >= 4-fold rise from baseline.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.
Two-sided 95% (or one-sided 97.5% for percentages of 0% or 100%) CIs for percentages are presented using Clopper-Pearson method.

B1222, Participants receiving a third dose booster of AZD1222; CI, Confidence interval; GMT, Geometric mean titre; LLoQ, Lower limit of quantification; n, Number of subjects in analysis; N, Number of subjects per treatment group; NE Not Evaluable; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222
Source: Tables 14.2.1.1 and see Appendix E, IEMT Table 19.2.1.1

Conclusion

In totality, these data show that GMTs and corresponding GMT ratios were not overly influenced by a small number of high-responding study participants and indeed reflect the ability of an AZD1222 boost to counteract the effects of waning immunogenicity after primary series vaccination. Moreover, these analyses further confirm that lower seroresponse rates observed post booster as compared to primary series vaccination are reflective of higher baseline titres rather than an abnormal distribution of immunological responses within boosted study participants.

Assessment of MAH's response:

The MAH has provided the additional analyses requested. In particular, the MAH provided histograms [(showing the percentage of subjects reaching different antibody titres (pre- and post- booster)], reverse cumulative distribution curves (RCDC) of pseudoneutralising antibodies (pre- and post- booster doses), and a specific analysis of the booster responses in participants with antibody titres pre-booster bellow the LLOQ. In most cases, data are provided based both in terms of pseudovirus neutralizing antibodies and S-protein binding antibodies.

Overall, the histogram data and the RCDCs for both the V1222/B1222 and the VmRNA/B1222 cohorts do not indicate that there are two different subpopulations, one of high responders and the other responding poorly to the booster dose. Rather, the data suggest that most of the subjects that received a booster dose experienced an increased in nAb titres.

It is noted that the RCDCs (based on Wuhan pseudoneutralising antibodies) of the cohorts V1222/B1222 and the historical control group (HV1222)(HCG) (Figure 2) practically overlap, a result that agrees well with the GMTR of 1.03 (95%CI 0.97-1.14) observed for these two cohorts. These data indicate, as already mentioned in the previous assessment report, that the AZD1222 booster dose restores in the AZD1222 previously vaccinated cohort the titres reached after a primary vaccination series of AZD1222.

For both the V1222 and VmRNA groups, at day 29 after AZD1222 booster, a higher rate of seroresponse (81.4% and 97.3%, respectively) was observed in participants with baseline nAb titres < LLOQ than in the overall population [66.1% (V1222) and 43.2% (VmRNA)] (Table 2). It is noted that in the HCG, the seroresponse rate after primary vaccination was 84.1%. Thus, the submitted dataindicate that AZD1222 injection strongly boosts the immune response in subjects from the VmRNA and V1222 groups with titres bellow the LLOQ. The lower seroresponse observed in the overall population from both groups is then a consequence of the difficulty of achieving a ≥ 4-fold rise in nAb titres in subjects with high titres before receiving the booster dose.

In conclusion, the data provided do not indicate that meeting the primary endpoint (based on GMTs ratio) for both the V1222/B1222 and VmRNA/B1222 were due to a small number of high-responding study participants. Rather, the data indicate that most of the subjects increased the nAb titres after the AZD1222 booster, although high seroconversion rates were observed in subjects with pre-booster titres below the LLOQ since it was more difficult to achieve seroresponse of ≥ 4-fold rise from baseline in subjects with higher pre-booster titres. This is a general observation made also for many other vaccines.

The MAH is asked to consider including data on the seroresponse rate achieved after a booster dose in the SmPC.

Conclusion: Point Solved.

Question 1c: Non-inferiority comparisons for boosting of mRNA vaccinated participants

Justification for not following the CHMP recommendation on the immunogenicity analysis to allow use of AZD1222 as booster for those previously vaccinated with an mRNA vaccine. The MAH did not follow the recommendation made by the CHMP on the comparisons to be made to support a claim for AZD1222 to be used to boost the response in persons previously vaccinated with COVID-19 mRNA vaccines. Instead, the MAH compared the response following AZD1222 booster to the historical control group that received two doses of

AZD1222. The validity of this approach is questioned in that reaching the primary endpoint is not a demanding requirement, considering that the baseline GMT of the VmRNA/B1222 group was 197 (95% CI 179-271)] which was quite close to the GMT reached after primary vaccination in subjects from the historical control group [242 (95% CI 224-262)]. Moreover, the fact that only 43% of the boosted subjects seroconverted, questions the claim that AZD1222 can be used to boost subjects that received a primary vaccination with an mRNA vaccine. The MAH is asked to comment.

Summary of MAH's Response:

AstraZeneca was unable to follow the CHMPs recommendation regarding an mRNA primary series treatment group. It was not possible to access serum samples for the VmRNA cohort recruited to the trial in order to assess immunogenicity at Day 29 following a primary series. Nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study.

It is acknowledged that the previously mRNA vaccinated participants had higher neutralising antibodies at baseline than the previously AZD1222 vaccinated participants (eg, GMT of 198 vs 38 for nAbs against Wuhan-Hu-1). Despite this relatively high baseline level, a booster of AZD1222 in VmRNA participants resulted in a 3.77-fold increase to a GMT of 747, compared to a GMT of 249 reached in the V1222 cohort. Both these levels exceeded the value associated with clinical protection following primary series vaccination (ie, GMT 174.8 [159.7, 191.2]; Table 33 in Module 2.7.3, dated 25 February 2021, eCTD sq 18).

Further, while overall in the mRNA cohort there was a 3.77-fold increase in GMTs, in participants with lower baseline nAbs there was a greater increase in nAbs (as discussed in the response to Query 1b above). In mRNA cohort participants with nAbs < LLOQ at baseline, there was a 27.12-fold increase in GMTs to 542.5 and 97.3% seroresponse. In summary, while higher baseline GMTs impede the ability to demonstrate seroresponse, participants with both low and high baseline GMTs experience a booster response to levels associated with clinical protection, with the magnitude of the response being greater in those with lower baseline levels.

Assessment of MAH's response:
The MAH acknowledges that it has not followed the CHMP advice to compare the immune response of the VmRNA/B1222 group to an mRNA primary series treatment group. The MAH indicates that it was not possible to access serum samples for the VmRNA cohort nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study.

On the other hand, the MAH notes that a booster of AZD1222 in VmRNA participants resulted in a 3.77-fold increase to a GMT of 747, with this level exceeding the value associated with clinical protection following primary series vaccination (ie, GMT 174.8 [159.7, 191.2]. Moreover, as noted in the response to Query 1b (above), in mRNA cohort participants with nAbs < LLOQ at baseline, there was a 27.12-fold increase in GMTs to 542.5 and 97.3% seroresponse (Table 2).

Taking into account all these observations (in particular the figure of 3.08 of the GMTR -VmRNA/B1222 vs HCG- and the high seroresponse rate in participants with nAbs < LLOQ at baseline), it is considered that the approach followed by the MAH would support the use of AZD1222 as heterologous boost in subject that received a primary vaccination series with an mRNA vaccine authorized in the EU. It is noted, however, that the clinical data supporting this heterologous booster should be clearly stated in the SmPC.

Conclusion: Point solved.

Conclusion

☒Overall conclusion and impact on benefit-risk balance has/have been updated accordingly

9.2. Other concerns

Clinical aspects

Question 2: Dosing interval of historical control group compared with booster groups

In the document submitted to EMA by the MAH seeking advice on switching to use an historical control as the comparator arm for the previously vaccinated treatment groups it was stated: "The inclusion/exclusion criteria of D7220C00001 and D8110C00001 are similar, and the data will not be confounded by dosing interval and the dose level, unlike the AZD1222 studies conducted by University of Oxford which included subjects who were vaccinated with 2 doses at various intervals (4 weeks to 16 weeks) and a subset of participants who were administered a low dose of AZD1222. […] Given that the dosing interval in the previously vaccinated group in the D7220C00001 study is 4 weeks, using data from studies with longer dosing interval is not appropriate in a trial designed to evaluate non-inferiority."

In our understanding this text stated that the dosing interval (for the primary series of AZD1222) in the previously vaccinated group from trial D7220C00001 was going to be the same as in trial D8110C00001, which used an interval of 4 weeks. However, according to the data submitted by the MAH, the dosing interval in the previously AZD1222- vaccinated group in the study D7220C00001 varied from 25 to 91 days with a median of 59 days. The same situation applies to the previously vaccinated mRNA cohort (the interval between first and second dose ranged from 21 to 86 days with a median of 70 days). Moreover, as indicated above, the MAH stated that "using data from studies with longer dosing interval is not appropriate in a trial designed to evaluate non-inferiority", a sentence that questions the approach followed for the comparisons made in this variation application. The MAH is asked to comment on these statements.

Summary of MAH's Response:

The statements regarding the similarities of inclusion/exclusion criteria for studies D7220C0001 and D8110C00001 were intended to apply to comparisons of the control cohort (ie, the vaccine-naïve population in D7220C00001, which was proposed to be replaced as the control cohort by a matched population from study D8110C00001). In this respect, the 4-week dosing interval in D8110C00001 matched with the planned dosing interval for the primary series control cohort in D7220C00001.

It was not expected that the primary series dosing interval for the booster cohorts in D7220C00001 would match with the control cohort. As originally designed, the vaccine-naïve control cohort in D7220C00001 was to receive primary series doses at a 4-week interval. Conversely, as per inclusion criterion 8, the previously-vaccinated cohort in D7220C00001 was to have received 2 doses of AZD1222 or mRNA-1273 at a 4- to 12-week dosing interval or 2 doses of BNT162b2 at a 3- to 12-week dosing interval, in line with dosing authorisations/recommendations in the countries where the D7220C00001 study was conducted.

The decision to use data from study D8110C00001 for the historical control group was informed by many factors beyond primary series dosing interval. These included broad similarities in inclusion/exclusion criteria between the studies as well as a large proportion of elderly participants in study D8110C00001 (whereas the University of Oxford studies included few elderly participants with available pseudoneutralising antibody data).

Importantly, the differences in primary series vaccination interval have not confounded the results.

Sensitivity analysis of immunogenicity including model term for interval between the 2 primary series doses confirm the results of the primary analysis (full results were previously provided in a Supplementary Sensitivity Analyses of Study D7220C00001 Interim Results dated 24 February 2022). As noted in the response to Query 3, below, a trend of higher immunogenicity with shorter primary series intervals was observed (Table 4 and Table 5). Consequently, the use of this historical control data set led to a more stringent criteria for demonstrating non-inferiority.

Assessment of MAH's response:

The explanation provided by the MAH is considered satisfactory.

Conclusion: Point solved.

Question 3: Sub-group immunogenicity by primary vaccination and booster dose intervals

Regarding the cohorts boosted with AZD1222, there is a wide time interval between first and second doses (see question 2), and also between second and booster dose (for the AZD1222 cohort: 74 to 379 days with a median of 269 days; and for the mRNA cohort: 73 to 213 days with a median of 123 days). The MAH is asked to carry out a subgroup analysis to determine the impact of the dose interval in primary vaccination as well as the time interval from second to booster dose on the GMT and seroresponse rates reached.

Summary of MAH's Response:

Table 4 and Table 5 present immunogenicity data by primary series dose interval. While these results show the greatest fold rise in pseudoneutralising antibody titres against Wuhan-Hu-1 and Beta after AZD1222 booster dose were among participants with an interval of less than 6 weeks for both the AZD1222- and mRNA-vaccinated participants, these results reflect the lower baseline GMTs observed in this interval.

Table 6 and Table 7 present immunogenicity data by booster dose interval. These tables show that a booster dose of AZD1222 increases pseudoneutralising antibody titres against Wuhan-Hu-1 and Beta across all booster intervals though, as expected, the fold-rise was lower the participants with higher baseline GMTs (ie, the 3-6 month post-primary series groups). The GMTs against the Wuhan-Hu-1 strain reached levels considered to provide protection for all treatment intervals (ie, V1222 cohort: 188.89, 310.38, 273.14 for the < 6, 6-9, and > 9 month intervals, respectively; VmRNA cohort: 723.14 and 1198.46 for the < 6 and 6-9 month intervals, respectively).

Overall, it is AstraZeneca's view that the dosing interval between primary series and booster is a greater contributor to post-booster immunogenicity than the primary series interval itself. For a justification of AstraZeneca's proposal for setting the time between primary series and booster vaccination at ≥ 3 months, see the response to Query 12.

Note: For the subgroup analyses below, and other subgroup analyses presented in this response document, raw values are provided due to potential imbalances in prognostic factors within each subgroup. Noting that there have been only minor differences between raw and model-adjusted values, only raw values are presented in this response document so as to not complicate the responses with separate models and model estimates.

Table 4 **GMT, GMFR and seroresponse in V1222/B1222 participants for pseudoneutralising antibodies by dosing interval of primary series of AZD1222 (Seronegative immunogenicity analysis set)**

Statistic	<6 weeks			6 to <9 weeks			9-<12 weeks			≥12 weeks		
	Baseline N = 125	Day 29	Fold rise	Baseline N = 68	Day 29	Fold rise	Baseline N = 130	Day 29	Fold rise	Baseline N = 19	Day 29	Fold rise
Wuhan-Hu-1												
n	125	120	120	68	66	66	130	124	124	19	17	17
GMT	26.40	329.26	12.83	37.56	204.14	5.33	51.40	204.62	3.98	46.37	204.16	4.58
95% CI	(22.45, 31.06)	(271.25, 399.67)	(10.36, 15.90)	(27.89, 50.57)	(152.34, 273.54)	(3.75, 7.59)	(40.46, 65.29)	(159.74, 262.11)	(3.10, 5.12)	(24.76, 86.86)	(111.50, 373.84)	(2.49, 8.43)
Seroresponse % (n^a/N^b) (95% CI)	--	87.5 (105 / 120) (80.2, 92.8)		--	57.6 (38/66) (44.8, 69.7)		--	51.6 (64 / 124) (42.5, 60.7)		--	52.9 (9 / 17) (27.8, 77.0)	
Beta												
n	125	120	120	68	66	66	130	124	124	19	17	17
GMT	21.78	328.32	15.02	27.64	172.85	6.19	31.84	141.12	4.45	32.57	253.13	7.34
95% CI	(20.02, 23.71)	(260.41, 413.95)	(11.84, 19.05)	(22.54, 33.90)	(118.31, 252.52)	(4.05, 9.47)	(26.57, 38.17)	(107.74, 184.85)	(3.41, 5.80)	(20.37, 52.08)	(133.34, 480.53)	(3.30, 16.33)
Seroresponse % (n^a/N^b) (95% CI)	--	86.7 (104 / 120) (79.3, 92.2)		--	59.1 (39 / 66) (46.3, 71.0)		--	50.0 (62 / 124) (40.9, 59.1)		--	64.7 (11 / 17) (38.3, 85.8)	
Delta												
n	64	58	58	12	12	12	18	16	16	5	5	5
GMT	28.21	85.02	2.98	25.00	84.73	3.39	26.19	51.80	1.97	25.00	62.87	2.51
95% CI	(23.13, 34.40)	(68.34, 105.78)	(2.23, 3.98)	(NE, NE)	(51.96, 138.18)	(2.08, 5.53)	(23.74, 28.91)	(37.48, 71.58)	(1.42, 2.72)	(NE, NE)	(21.32, 185.40)	(0.85, 7.42)
Seroresponse % (n^a/N^b) (95% CI)		37.9 (22 / 58) (25.5, 51.6)			41.7 (5 / 12) (15.2, 72.3)			18.8 (3 / 16) (4.0, 45.6)			40.0 (2 / 5) (5.3, 85.3)	

[a] n is the number of seroresponders.
[b] N is the number of subjects with a baseline measurement and an assessment at the given time point.
Seroresponse is defined as a >= 4-fold rise from baseline.
Titre values measured as below LLoQ (40) are imputed to a value that is half of the LLoQ. Titre values measured as above ULoQ (787,339) are imputed at the ULoQ value.
GMT is calculated as the antilogarithm transformation of the mean of the log-transformed titre.
Two-sided 95% (or one-sided 97.5% for percentages of 0% or 100%) CIs for percentages are presented using Clopper-Pearson method.
CI confidence interval; GMFR geometric mean fold rise; GMT geometric mean titre; n Number of subjects in analysis;
N Number of subjects per treatment group and dosing interval; SD Standard deviation; NE Not Evaluable;
LLoQ Lower limit of quantification; ULoQ Upper limit of quantification; V primary vaccination; B booster vaccination.
Source: Appendix E, IEMT Table 15.2.1.1

Table 5 **GMT, GMFR and seroresponse in VmRNA/B1222 participants for pseudoneutralising antibodies by dosing interval of primary series of mRNA vaccine (Seronegative immunogenicity analysis set)**

Statistic	<6 weeks			6 to <9 weeks			9-<12 weeks			≥12 weeks		
	Baseline N = 47	Day 29	Fold rise	Baseline N = 23	Day 29	Fold rise	Baseline N = 216	Day 29	Fold rise	Baseline N = 8	Day 29	Fold rise
Wuhan-Hu-1												
n	47	46	46	23	22	22	216	202	202	8	8	8
GMT	115.98	1240.76	11.11	286.91	953.90	3.44	315.54	1002.87	3.07	464.08	834.42	1.80
95% CI	(76.79, 175.17)	(956.53, 1609.45)	(7.54, 16.35)	(165.31, 497.96)	(639.07, 1423.81)	(1.82, 6.49)	(265.33, 375.26)	(873.06, 1151.98)	(2.65, 3.56)	(112.53, 1913.93)	(449.15, 1550.18)	(0.61, 5.29)
Seroresponse % (n^a/N^b) (95% CI)	--	78.3 (36 / 46) (63.6, 89.1)		--	36.4 (8 / 22) (17.2, 59.3)		--	36.1 (73 / 202) (29.5, 43.2)		--	37.5 (3 / 8) (8.5, 75.5)	
Beta												
n	47	46	46	23	22	22	216	202	202	8	8	8
GMT	55.86	1042.35	19.53	117.87	549.13	4.76	140.66	714.57	4.86	329.68	573.65	1.74
95% CI	(39.37, 79.25)	(707.79, 1535.05)	(13.01, 29.32)	(71.20, 195.13)	(332.19, 907.76)	(2.65, 8.55)	(116.01, 170.55)	(606.10, 842.46)	(4.14, 5.71)	(92.88, 1170.19)	(291.33, 1129.54)	(0.77, 3.94)
Seroresponse % (n^a/N^b) (95% CI)	--	87.0 (40 / 46) (73.7, 95.1)		--	45.5 (10 / 22) (24.4, 67.8)		--	54.0 (109 / 202) (46.8, 61.0)		--	12.5 (1 / 8) (0.3, 52.7)	
Delta												
n	47	19	19	23	3	3	69	57	56	2	2	2
GMT	35.98	279.91	8.03	113.29	129.82	1.15	67.68	156.88	2.44	120.50	197.21	1.64
95% CI	(25.70, 50.35)	(193.26, 405.41)	(5.00, 12.89)	(62.06, 206.80)	(75.65, 222.76)	(1.06, 1.24)	(55.25, 82.92)	(126.73, 194.21)	(1.94, 3.08)	(17.84, 814.01)	(5.77, 6742.01)	(0.01, 377.95)
Seroresponse % (n^a/N^b) (95% CI)	--	84.2 (16 / 19) (60.4, 96.6)		--	0 (0 / 3) (0, 76.8)		--	25.0 (14 / 56) (14.4, 38.4)		--	0 (0 / 2) (0, 88.8)	

[c] n is the number of seroresponders.

Table 6 **GMT, GMFR and seroresponse in V1222/B1222 participants for pseudoneutralising antibodies by booster dose interval dose (Seronegative immunogenicity analysis set)**

	Booster dose interval <6 months (N = 112)			Booster dose interval 6-9 months (N = 71)			Booster dose interval >9 months (N = 159)		
	Baseline	Day 29	Fold rise	Baseline	Day 29	Fold rise	Baseline	Day 29	Fold rise
95% CI	(53.19, 93.43)	(145.23, 247.01)	(2.18, 3.50)	(22.25, 36.58)	(239.69, 436.12)	(7.98, 15.60)	(23.94, 31.32)	(213.47, 305.78)	(7.61, 11.51)
Seroresponse rate (%) 95% CI	--	40.6 (43 / 106) (31.1, 50.5)		--	79.4 (54 / 68) (67.9, 88.3)		--	77.8 (119 / 153) (70.4, 84.1)	
Beta									
n	112	106	106	71	68	68	159	153	153
GMT	40.66	91.67	2.23	23.94	375.57	15.57	21.33	278.14	13.01
95% CI	(32.45, 50.95)	(69.29, 121.29)	(1.77, 2.82)	(20.44, 28.04)	(274.24, 514.34)	(11.22, 21.60)	(20.10, 22.63)	(225.48, 343.10)	(10.43, 16.22)
Seroresponse rate (%) 95% CI	--	31.1 (33 / 106) (22.5, 40.9)		--	85.3 (58 / 68) (74.6, 92.7)		--	81.7 (125 / 153) (74.6, 87.5)	
Delta									
n	3	3	3	27	21	21	69	67	67
GMT									
95% CI									
Seroresponse rate (%) 95% CI	--	33.3 (1 / 3) (0.8, 90.6)		--	38.1 (8 / 21) (18.1, 61.6)		--	34.3 (23 / 67) (23.2, 46.9)	

Seroresponse is defined as a >4-fold rise from baseline.

Table 7 **GMT, GMFR and seroresponse in VmRNA/B1222 participants for pseudoneutralising antibodies by booster dose interval (Seronegative immunogenicity analysis set)**

	Booster dose interval <6 months (N = 276)			Booster dose interval 6-9 months (N = 18)			Booster dose interval >9 months		
	Baseline	Day 29	Fold rise	Baseline	Day 29	Fold rise	Baseline	Day 29	Fold rise
Wuhan-Hu-1									
n	276	260	260	18	18	18	--	--	--
GMT	290.65	1023.73	3.46	85.71	1112.37	12.98	--	--	--
95% CI	(248.13, 340.45)	(908.80, 1153.21)	(3.00, 4.00)	(43.39, 169.32)	(732.69, 1688.80)	(6.71, 25.10)	--	--	--
Seroresponse rate (%) (95% CI)	--	40.8 (106 / 260) (34.7, 47.0)		--	77.8 (14 / 18) (52.4, 93.6)		--		
Beta									
n	276	260	260	18	18	18	--	--	--
GMT	129.71	734.65	5.53	50.92	826.25	16.22	--	--	--
95% CI	(109.58, 153.54)	(635.69, 849.01)	(4.73, 6.46)	(28.16, 92.10)	(402.95, 1694.23)	(7.50, 35.10)	--	--	--
Seroresponse rate (%) (95% CI)	--	56.5 (147 / 260) (50.3, 62.7)		--	72.2 (13 / 18) (46.5, 90.3)		--	--	
Delta									
n	95	81	80	--	--	--	--	--	--
GMT	60.56	179.46	3.12	--	--	--	--	--	--
95% CI	(50.90, 72.04)	(150.09, 214.56)	(2.48, 3.92)	--	--	--	--	--	--
Seroresponse rate (%) (95% CI)	--	37.5 (30 / 80) (26.9, 49.0)		--	--		--	--	

Seroresponse is defined as a ≥4-fold rise from baseline.

Assessment of MAH's response:

The MAH has submitted the data requested. It is not obvious why in Tables 4 (V1222/B1222) and 5 (VmRNA/B1222), in general, the nAB GMT reached after the booster doses were higher in those that received the primary vaccination series with the shortest time intervals (<6 weeks) as compared to the groups for which the primary dose interval was higher. It was not clear either why the baseline titres increase as long as the time intervals between the doses get longer. Having said that, the confidence intervals of these calculated GMT (according to various primary dosing intervals) were wide, and thus no obvious effect of the primary series dose interval on the nAb GMT reached after AZD1222 booster was observed.

The results shown in Table 6 (V1222/B1222) and Table 7 (VmRNA/B1222) do not show a clear effect of

different booster dose intervals on the GMT reached after booster dose.

In conclusion, these data do allow the use of homologous or heterologous booster dose according to the primary dosing intervals stated in the current SmPCs of Vaxzevria and the mRNA vaccines, respectively. Moreover, the data provided indicate that no negative effect is observed if this time interval is longer. The data on the booster dose intervals are discussed later in query 12.

Conclusion: Point solved.

Question 4: Inclusion of participants with booster dose intervals of 70 to 89 days

The MAH states that: "Following a blinded review of protocol deviations related to the timing of doses, it was decided that only participants with intervals of <70 days from second dose to booster dose … would be excluded from the immunogenicity analysis sets." Considering that usually for vaccines, booster doses are administered at 3-6 months after last vaccination, the MAH should provide the rationale for including participants with less than 90 days from second dose to booster dose in the analysis.

Summary of MAH's Response:

The inclusion criteria for study D7220C00001 stipulated there was to be a minimum of 90 days between previous vaccination and administration of study intervention (ie, booster dose). There were 4 previously vaccinated seronegative participants who received the booster dose fewer than 70 days after their second dose. These participants were excluded from the Seronegative Immunogenicity Analysis Set. There were 13 seronegative participants who received the booster dose 70 days or more but fewer than 90 days after their second dose. Assuming that immunogenicity halves every 6 months and that the decay follows an exponential distribution, the difference between Day 70 and Day 90 would be approximately 5.7%. Given this modest change in immunogenicity relative to the gain in precision in estimating immunogenicity post-booster by maximizing the number of participants contributing data, a decision was made prior to database lock and documented in the PDMP to include participants with an interval of less than 90 days in the Seronegative Immunogenicity Analysis Set. Moreover, given that the primary analysis of the data included a term for time since previous vaccination, this was accounted for in the model-adjusted analysis. The similarity in results between raw and model-adjusted data further support the rationale of including these subjects.

The immunogenicity assessment presented in the CSR has been re-analysed with data that includes only those participants with an interval of 90 days or greater between the second dose and the booster dose. Table 8 and Table 9 demonstrate very similar immunogenicity at Day 15 and Day 29 in previously vaccinated participants with intervals of 90 days or greater and 70 days or greater, respectively, between the second dose of primary series of AZD1222 and an AZD1222 booster dose (GMTs against Wuhan-Hu-1 of 244.90 vs. 243.50 at Day 29 in the ≥ 90 and > 70 analyses, respectively). Similar results were observed in participants previously vaccinated with an mRNA vaccine (Table 10 and Table 11; GMTs: 1024.37 vs. 1029.25, respectively). Notably, in the V1222 cohort, only 4 of the 327 participants in the Day 29 Wuhan-Hu-1 and Beta analyses and 1 of the 90 participants in the Day 29 Delta analysis had a dosing interval of less than 90 days. These Four Tables (8 to 11) are not included in this summary.

Assessment of MAH's response:

The MAH explains that the decision to incorporate the 13 seronegative participants, who received the booster dose 70 days or more but fewer than 90 days after their second dose, was to gain in precision in estimating immunogenicity post-booster by maximizing the number of participants contributing data.

The MAH provides now Tables, which show very similar immunogenicity results in previously vaccinated participants with intervals of 90 days or greater and 70 days or greater, between the second dose of

primary series of AZD1222 and an AZD1222 booster dose.

In conclusion, the incorporation in the seronegative immunogenicity analysis set of subjects who received the booster dose booster dose 70 days or more but fewer than 90 days after their second dose does not show any significant impact in the immunogenicity results obtained.

Conclusion: Point solved (see also query 5).

Question 5: Seronegative participants with booster dose interval of 70 to 89 days

In total there were 13 seronegative participants who received the booster dose ≥ 70 days but < 90 days after their second dose, who were included in the Seronegative Immunogenicity Analysis Set. The MAH is asked to clarify whether these 13 subjects received a booster dose of AZD1222 or AZD2816.

Summary of MAH's Response:

Of the 13 participants included in the Seronegative Immunogenicity Analysis Set who received the booster dose 70 to 89 days after their second primary vaccination dose, 4 participants were in the V1222/B1222 treatment group, 5 participants were in the VmRNA/B1222 treatment group, and 4 participants were in the VmRNA/B2816 treatment group.

Assessment of MAH's response:

The MAH has provided the information requested.

Conclusion: Point solved (see also query 4).

Question 6: Seroconversion rate based on >2-fold increase in S-binding antibodies.

Based on the precision of the assay for detection of S-binding antibodies (Multiplexed ECL Method for the Detection of SARS-CoV-2 S) against the Wuhan strain, the MAH is asked to discuss the reliability of the data on seroconversion rate (based on a ≥2-fold increase) to assess the seroconversion rate achieved by the AZD1222 booster. If the MAH considers that the reliability is adequate, the MAH is asked to provide a Table with the percentage of seroresponders (≥2-fold) against the Wuhan strain in the cohorts boosted with AZD1222 and calculate accordingly the serosresponse rate as compared to the historical control group.

Summary of MAH's Response:

Based on the previously-reported intermediate precision estimate of 20.1% geometric coefficient of variation for the ancestral Spike protein on the multiplexed ECL based Spike assay (see Module 2.7.1, Table 4), there is a > 99% power to detect a ≥ 2-fold rise in Spike-binding antibodies for all relevant observed sample sizes. Given the strength of this power assessment, AstraZeneca believe it is appropriate to calculate seroresponse based on a ≥ 2-fold rise in Spike-binding antibodies.

Table 12 and Table 13 provide seroresponder analyses where seroresponse is defined as a ≥ 2-fold rise from baseline for nAbs and spike-binding antibodies, respectively. While increases in seroresponse rate were observed as compared to the ≥ 4-fold rise criteria, seroresponse was still lower than that observed for primary series AZD1222 vaccination.

Table 12 **Seroresponse Comparisons (defined as ≥ 2-fold rise from baseline at Day 29) for Wuhan-Hu-1 Pseudoneutralising Antibodies (Seronegative Immunogenicity Analysis Set)**

	Statistic	Comparator	Reference (Historical control)	Seroresponse difference
V1222/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	342	508	327/508
	Value % (n[a]/N[b])	75.5 (247 / 327)	89.2 (453 / 508)	-13.6
	95% CI	70.5, 80.1	86.1, 91.7	-19.1,-8.4
VmRNA/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	294	508	327/508
	Value % (n[a]/N[b])	66.2 (184 / 278)	89.2 (453 / 508)	-23.0
	95% CI	60.3, 71.7	86.1, 91.7	-29.2,-16.9

[a] n is the number of seroresponders.
[b] N is the number of subjects with a baseline measurement and an assessment at the given time point.
Seroresponse is defined as a >= 2-fold rise from baseline.
Data are Day 29 post-booster for the Comparator group and Day 29 post-dose 2 for the Reference group.
The population of historical controls is selected from those who had baseline and Day 29 post dose 2 pseudoneutralisation data.
Two-sided 95% (or one-sided 97.5% for percentages of 0% or 100%) CIs for percentages are presented using Clopper-Pearson method.
The difference in seroresponse is calculated as (seroresponse rate of comparator arm) - (seroresponse rate of reference arm), where the seroresponse rate is calculated from those who have titre assessments at baseline and the time point of interest. The CI for the difference in seroresponse is calculated using the Newcombe method based on the Wilson score
B1222, Participants receiving a third dose booster of AZD1222; CI, Confidence interval; n, Number of subjects in analysis; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222
Source: see Appendix E, IEMT Tables 20.1 and 20.2

Table 13 **Seroresponse Comparisons (defined as ≥ 2-fold rise from baseline at Day 29) for Wuhan-Hu-1 Spike Protein Binding Antibodies (Seronegative Immunogenicity Analysis Set)**

	Statistic	Comparator	Reference	Seroresponse difference
V1222/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	342	508	321/508
	Value % (n[a]/N[b])	82.6 (265 / 321)	99.2 (504 / 508)	-16.7
	95% CI	78.0, 86.5	98.0, 99.8	-21.2, -12.7
VmRNA/ B1222 (Wuhan-Hu-1) vs. Historical Control (Wuhan-Hu-1)	n	294	508	270/508
	Value % (n[a]/N[b])	71.1 (192 / 270)	99.2 (504 / 508)	-28.1
	95% CI	65.3, 76.4	98.0, 99.8	-33.8, -22.9

[a] n is the number of seroresponders.
[b] N is the number of subjects with a baseline measurement and an assessment at the given time point.
Seroresponse is defined as a >= 2-fold rise from baseline.
Data are Day 29 post-booster for the Comparator group and Day 29 post-dose 2 for the Reference group.
The population of historical controls is selected from those who had baseline and Day 29 post dose 2 spike protein binding data.
Two-sided 95% (or one-sided 97.5% for proportions of 0% or 100%) CIs for proportions are presented using Clopper-Pearson method.
The difference in seroresponse is calculated as (seroresponse rate of comparator arm) - (seroresponse rate of reference arm), where the seroresponse rate is calculated from those who have titre assessments at baseline and the time point of interest. The CI for the difference in seroresponse is calculated using the Newcombe method based on the Wilson score
B1222, Participants receiving a third dose booster of AZD1222; CI, Confidence interval; n, Number of subjects in analysis; VmRNA, Participants previously vaccinated with 2 doses of an mRNA vaccine; V1222, Participants previously vaccinated with 2 doses of AZD1222
Source: see Appendix E, IEMT Tables 20.2.1 and 20.2.2

Assessment of MAH's response:

The MAH has provided the information requested.

It is agreed with the MAH that the good intermediate precision of assay measuring Spike- binding antibodies (the multiplexed ECL based Spike assay) makes it relevant to calculate seroresponse rates based on a ≥ 2fold rise in Spike-binding antibodies to shed light on the immune response raised after

AZD1222 booster dose.

The seroresponse rates determined when using a ≥ 2fold rise in binding antibodies were 82.6% (95%CI 78- 86), and 71.1% (95%CI 65-76) for the V1222/B1222 and the VmRNA/B1222 cohorts, respectively. These figures significantly increased from those calculated when using a ≥ 4fold rise in binding antibodies: 68.2% (95%C 62-73) for the V1222/B1222 and 36.7% (95%CI 30-42) for the VmRNA/B1222. The seroresponse rate was practically unaltered for the Historical control group 98.8% [(95%CI 97-99) to 99.2% (95%CI 98-99)].

The results based on a ≥ 2-fold rise in binding antibodies have also been described in other studies aimed at measuring the immunogenicity reached following a homologous or heterologous boost with COVID-19 vaccines (e.g., Atmar et al. DMID 21-0012 Study Group. N. Engl. J. Med. 2022; 386:1046-1057). In the context of this trial, the data obtained are interpreted in that the seroresponse rate in terms of S-binding antibodies (≥ 2-fold rise) is quite significant for both cohorts (V1222/B1222 and VmRNA/B1222), and thus these data indicate that the AZD1222 booster dose is in fact boosting the response induced after primary vaccination in most of the participants in the trial.

Certainly, the seroresponse rates reached are still lower than those achieved after primary vaccination series in the HCG. It is also noted that the relevance of S-binding antibodies for clinical protection is unclear, but a number of studies have shown that S- binding and nAb titres correlate, and thus it an increase in protection in subjects with only a 2-fold increase in S-binding antibodies is expected. Despite these limitations, the data described in this query together with those based on seroresponse rates of nAb in subjects with antibody titres below the LLOQ before booster dose (see query 1b) are interpreted in that the AZD1222 booster dose increases nAb antibody titres in a large proportion of the subjects receiving the AZD1222 booster. This interpretation was not so obvious when using seroresponse rates based on nAb in the overall populations of the two cohorts or when using on a ≥ 4fold rise of S-binding antibodies.

In conclusion, all the data described above provide support for the use of AZD1222 as a homologous or heterologous boost.

Conclusion: Point solved.

Question 7: CSR Tables 14 and 15 footnote clarification

In Tables 14 and 15 (interim CSR) it is indicated under superscript "a" that baseline titre for reference cohort: "For historical control, values at Day 29 after the first dose of the primary series (ie, prior to administration of the second dose of the primary series)". It is understood that this is a mistake and that the MAH is referring to values pre-dose 1. The MAH is asked to clarify.

Summary of MAH's Response:

The Rapporteur is correct. For CSR Table 14, the historical control 'baseline' data originally included in this table were data from Day 29 (pre-second dose). These data were then updated to pre-dose Day 1 measurements. However, the footnote was not changed to reflect the new data. Similarly, for Table 15 the footnote (and the data; see below) were erroneously not updated. For both the comparator and the reference (historical control) cohorts, the baseline antibody titres in CSR Tables 14 and 15 are intended to be those from pre-dose Day 1 measurements.

Assessment of MAH's response:

The MAH has clarified the question raised.

Conclusion: Point solved.

Question 8: CSR Tables 14 and 15 historical control baseline data

In Table 15 from interim CSR, the baseline data for the Reference group (historical control group) are different from those stated in Table 14 for this same group. It is understood that the correct ones are those from Table 14. The MAH is asked to clarify.

Summary of MAH's Response:

The Rapporteur is correct. The corrected CSR Table 15, including revised footnotes, has been provided (not included in this Assessment report)

Note that the Day 29 fold rises in the reference (historical control) group in the original table were calculated based on the correct Day 1 baseline results and thus are correct.

Assessment of MAH's response:

The MAH has clarified the question raised.

Conclusion: Point solved.

Question 9: Baseline characteristics of historical control group and AZD1222 booster groups

The MAH should provide a comparative table of the patients previously vaccinated with two doses of AZD1222 from the study D8110C00001 (historical control group) and the patients recruited in this study (V1222/B1222 and VmRNA/B1222) with the standardised differences of the baseline characteristics for each groups of interest. The p-values should also be provided but only for exploratory purposes.

Summary of MAH's Response:

See Query 11 (Table 14, Table 15) for demographic and baseline characteristics of the V1222/B1222, VmRNA/B1222, and historical control groups.

Assessment of MAH's response:

See response to query 11.

Conclusion: Point solved

Question 10: Study D8110C00001 full study and historical control group baseline and demographic characteristics

The MAH is asked to present a table comparing the differences in baseline and demographic characteristics between the subjects incorporated in the historical group from trial D8110C00001 and the subjects excluded from the historical group of the same trial. Additionally, another table with their standardised differences (including their p-values) should be provided.

Summary of MAH's Response:

Appendix E, IEMT Tables 506.2.1, 506.2.2, and 506.2.3, provide demographic and baseline characteristic details for eligible participants from study D8110C00001 included/not included in the matched historical control group analysis set.

The differences in the characteristics of participants included/not included in the historical control group were as expected. Age, sex, and the presence of at least 1 baseline comorbidity were matching criteria and participants in study D8110C00001 were younger, more likely to be male, had a higher rate of baseline comorbidities, and had higher baseline BMI than the study D7220C00001 participants previously vaccinated with AZD1222. By design, the matching algorithm selected controls who were similar to the V1222 cohort in study D7220C00001 for comparability.

Importantly, for the other baseline characteristics not included in the matching algorithm, including primary vaccination dosing interval, the characteristics were similar between the included/not included groups. Historical control group matching is further discussed in the response to Query 11, below.

Assessment of MAH's response:

The Applicant has presented a number of tables comparing the demographic and baseline characteristics of the patients' data used as historical control from the D8110C00001 trial against those patients' data who were not eligible for the matched historical control group analysis set.

From the tables presented, it can be stated that there are not great differences between groups. However, the Applicant was asked to present the standarised differences (STD) for these comparisons. Regrettably, the Applicant has not provided this information and it wold have been useful to assess the magnitude of the differences in this context. However, this issue not further pursued, and the query can be considered solved.

Conclusion: Point solved.

Question 11: Baseline and demographic characteristics of historical control group

The MAH provides a Table (Table 11**) with few details regarding the historical control group. The MAH is asked to provide Tables with baseline and demographic characteristics (similar to Tables 8 and 9 from the Interim CSR) with three columns corresponding to the V1222/B1222, VmRNA/B1222 and the historical control groups in order to assess the good match of the AZD1222 boosted cohorts with the historical control group. In case differences are found between the historical control group and the two other cohorts (V1222/B1222 and VmRNA/B1222), the MAH is asked to discuss their impact in regard to the primary and key secondary immunogenicity analysis.**

Summary of MAH's Response:

The demographic and baseline characteristics of the V1222/B1222, VmRNA/B1222, and historical control groups, including p-values, are provided in Table 14 and Table 15, respectively. This historical control group was selected through propensity score matching between a pool of all participants in the immunogenicity analysis set from study D8110C00001 with all participants previously vaccinated with AZD1222 in study D7220C00001. All matched participants from study D8110C00001 with available neutralising antibody data were then included in the historical control group.

The historical control and V1222/B1222 groups were well matched for key demographic and baseline characteristics, including age, sex, BMI, and presence of at least one comorbidity. As expected, there were more Hispanic and non-White participants in the historical control group, though race/ethnicity has not been shown to impact the efficacy, immunogenicity, or safety of AZD1222 and, as such, ethnicity was not included as a covariate for the one-to-one full matching algorithm. There was also a difference in primary vaccination dosing interval, with a median of 28 days in the historical control group, compared to 59 days in the V1222/B1222 group. This 28-day median dosing interval in the historical control group matches well with the 4-week interval for the originally planned vaccine naïve control group from study

D7220C00001.

As noted in the CSR (see Sections 10.4 and 13.1) there were differences in characteristics (notably age, sex, and time since prior vaccination) between the VmRNA/B1222 and V1222/B1222 groups. These differences also apply to a comparison between the VmRNA/B1222 and historical control groups. However, these differences were not considered to affect the validity of the results or the ability to reach conclusions. To assess the impact of these differences and allow for direct comparisons of AZD1222/AZD2816 booster dosing between the V1222 and VmRNA cohorts, model-adjusted immunogenicity analyses, and subgroup analyses of the immunogenicity and safety data, were conducted. The results of the model-adjusted analyses were consistent with the raw data results. Overall, the differences in participant characteristics between treatment groups were not considered to have a clinically meaningful impact on the primary and secondary immunogenicity analyses.

It can also be noted that the neutralising antibody GMTs in the historical control group were higher than those in the immunogenicity analysis set for the University of Oxford pooled analyses (ie, Wuhan-Hu-1 pseudoneutralising GMT = 224.82 vs 175.07 (SD/SD+LD/SD), respectively). This indicates that the selection of the historical subgroup through the appropriate propensity score matching from study D8110C00001 provided a higher bar than if data from University of Oxford studies were used as comparator. Nevertheless, non-inferiority was still met.

The primary analysis of immunogenicity used model adjusted values. The model adjustment estimated a regression coefficient for the impact of prognostic baseline characteristics for each treatment arm and then used the regression coefficients to standardise immunogenicity values to the mean level of the covariate range (ie, if male was coded as 0 and female coded as 1 then all participants were standardised to a sex of $0.5*\beta_{sex}$). The result of this are immunogenicity responses corrected for an imbalance in prognostic baseline characteristics between treatment arms. The fact that the non-inferiority conclusions reached were identical when using either the raw or model adjusted immunogenicity value support the notion that imbalances in baseline patient characteristics had a minimal impact on the observed immunogenicity results.

Characteristic/Statistic	V1222/ B1222 N = 342	VmRNA/ B1222 N = 294	Historical control N = 508
Age (years)			
Mean	59.7	55.5	59.4
Standard deviation	13.65	13.21	13.65
Median	62.0	55.0	62.0
P-value	0.8227 [a]	<0.0001 [a]	-
Age group, n (%)			
≥ 18 to < 65 years	185 (54.1)	215 (73.1)	273 (53.7)
≥ 65 years	157 (45.9)	79 (26.9)	235 (46.3)
P-value	0.9193	<0.0001	-
Sex, n (%)			
Male	186 (54.4)	112 (38.1)	266 (52.4)
Female	156 (45.6)	182 (61.9)	242 (47.6)
P-value	0.5620	<0.0001	-
Race, n (%)			
White	298 (87.1)	265 (90.1)	465 (91.5)
Black or African American	2 (0.6)	3 (1.0)	18 (3.5)
Asian	9 (2.6)	8 (2.7)	16 (3.1)
Mixed	0	2 (0.7)	5 (1.0)
Unknown	33 (9.6)	16 (5.4)	3 (0.6)
P-value	0.0127 [b]	0.0837 [b]	-
Ethnicity, n (%)			
Hispanic or Latino	6 (1.8)	3 (1.0)	34 (6.7)
Not Hispanic or Latino	295 (86.3)	268 (91.2)	460 (90.6)
Missing	41 (12.0)	23 (7.8)	14 (2.8)
P-value	0.0022	0.0004	-
Country, n (%)			
Poland	10 (2.9)	1 (0.3)	0
United Kingdom	332 (97.1)	293 (99.7)	0
USA	0	0	508 (100.0)
P-value	NA	NA	-

[a] The p-value assumes equal variances as the equality of variance assumption was not rejected.
[b] For race P-value calculation, categories are collapsed into 2 levels - White and Other (which includes all remaining race categories except Unknown)- to satisfy chi-square assumption that all expected cell counts >= 5.

P-values for continuous variables correspond to an independent t-test comparing the previously vaccinated participants against the historical control participants. A p-value < 0.05 gives evidence against equality.
P-values for categorical variables correspond to a chi-square test comparing the previously vaccinated participants against the historical control participants. A p-value <0.05 gives evidence against independence.
Missing and Unknown categories are not included in p-value calculations.
Percentages are based on N, the number of subjects in the analysis set for each treatment group.
Age groups are derived from the age groups specified at randomisation. Sex is derived from the corrected value after randomisation.
n Number of subjects in analysis for a continuous variable and number of subjects per category for a categorical variable; SD Standard deviation; V primary vaccination; B booster vaccination; NA Not applicable

Characteristic/Statistic	V1222/ B1222 N = 342	VmRNA/ B1222 N = 294	Historical control N = 508
Primary vaccination dosing interval (days) [a]			
n	342	294	508
Mean	54.2	63.4	37.6
Standard deviation	20.3	18.6	14.67
Median	59.0	70.0	28.0
Minimum	25	21	22
Maximum	91	86	80
P-value	<0.0001 [c]	<0.0001 [c]	-
BMI (kg/m^2)			
n	341	293	508
Mean	27.3	27.8	27.0
SD	5.2	5.8	4.83
Median	26.8	26.9	26.3
P-value	0.4277 [b]	0.0390 [c]	-
Comorbidity at baseline			
At least one comorbidity	158 (46.2)	138 (46.9)	237 (46.7)
BMI ≥ 30 kg/m^2	85 (24.9)	89 (30.3)	--
Significant cardiovascular disease	84 (24.6)	61 (20.7)	--
Chronic lung disease	30 (8.8)	27 (9.2)	--
Diabetes	20 (5.8)	9 (3.1)	--
None	184 (53.8)	156 (53.1)	271 (53.3)
P-value	0.8963	0.9378	-

[a] For previously vaccinated groups, the primary vaccination dosing interval refers to the primary series received prior to enrolment in the study; for the historical controls, this interval refers to the dosing interval observed in the D8110C00001 study.
[b] The p-value assumes equal variances as the equality of variance assumption was not rejected.
[c] The p-value assumes unequal variances as the equality of variance assumption was rejected at alpha = 0.05.

Assessment of MAH's response:

The MAH has provided the information requested.

As already discussed in the preliminary assessment report, the historical control and the V1222/B1222 group were well matched according to a number of baseline characteristics (age, sex, BMI, and presence of at least one comorbidity). The MAH explains now that race/ethnicity was not expected to impact the efficacy, immunogenicity, or safety of AZD1222 and, as such, ethnicity was not selected as matching criteria. This approach is agreed upon and thus the differences observed in race/ethnicity between the HCG and the booster cohorts are not expected to have an impact on the immunogenicity comparisons made. It is now clearly stated, as assumed in the preliminary assessment report, that the median primary vaccination dosing interval in the historical control group was 28 days, which is different from that of the V1222/B1222 (59 days) and VmRNA/B1222 (70 days) cohorts. As discussed above, in query 3, these differences in the primary dosing interval do not appear to have a significant impact on the immunogenicity comparisons made.

As also noted in the initial assessment report, there were differences in several characteristics (age, sex) between the VmRNA/B1222 and V1222/B1222 groups. The explanation provided by the MAH (the results of the model-adjusted analyses were consistent with the raw data results and the subgroup analyses of the immunogenicity) is considered to allow concluding that these differences do not have a clinically meaningful impact on the primary and secondary immunogenicity analyses.

It is noted that the Applicant was requested to provide the standardised differences (STD) (see Q9) between the three cohorts in order to better contextualise the difference in these comparisons. Regrettably, the Applicant hast not provided the STDs to assess the between-groups balance, and this

might have been really useful to assess the magnitude of those apparent differences. However, as stated by the Applicant, from a clinical point of view these differences does not seem to affect the validity of the results or the ability to reach conclusions so this issue is not further pursued.

Conclusion: Point solved.

Question 12: Justification for minimum time interval to receive the booster dose.

The MAH is asked to justify the proposed text in the SmPC that states "the third dose should be administered at least 3 months after completing the primary vaccination course" considering that the median time since second primary vaccination was 9 months in the AZD1222 cohort (range: 2.5 months to >1 year), and 4 months in the mRNA cohort (range: 2.5 to 7 months).

Summary of MAH's Response:

AstraZeneca considers that there are 2 main justifications for setting the time between primary series and booster vaccination at ≥ 3 months:

- Study D7220C00001 was designed to assess the immunogenicity and safety of booster doses administered ≥ 3 months after primary series and, based on data from the interim analysis, it was concluded that the study met its primary immunogenicity endpoint for this target population, and
- In the context of the ongoing pandemic and the emergence of more virulent variants, including those for which primary series vaccination with AZD1222 provides limited protection against infection (eg, Omicron), a broad booster vaccination window provides NITAGs with the flexibility to adapt their vaccination guidelines to the current local conditions.

The target population for the booster cohort in study D7220C00001 was participants previously vaccinated with AZD1222 or an mRNA vaccine at least 90 days prior to receiving study intervention. No upper boundary was set for time since primary series vaccination.

Of the 342 participants in the V1222/B1222 group included in the Seronegative Immunogenicity Analysis Set, 112 received their booster dose < 6 months after the primary series. For the 6-9 month and > 9 month intervals, the numbers were 71 and 159, respectively. As such, while the median interval in this group was 9 months, a substantial proportion of the participants who contributed to the primary immunogenicity endpoint (ie, 32.7%) had a dosing interval of between 2.5 and 6 months.

AstraZeneca acknowledges, as detailed in the response to Query 3, that GMTs after a booster dose of AZD1222 were lower in those who received the booster < 6 months from primary series compared with those with a longer interval between primary series and booster. However, these participants contributed to the overall positive outcome of the study. Further, while GMTs were comparatively lower, the booster dose in V1222/B1222 participants previously still increased GMTs by > 2x against each of the Wuhan, Beta, and Delta strains, and by > 3x in VmRNA/B12222 participants (see Table 6 and Table 7, respectively).

While a longer interval appears to result in a stronger booster response, setting the lower boundary at 3 months provides NITAGs with the flexibility to adapt their vaccination guidelines to the current local conditions. This was the position adopted in the UK, when on 09 September 2021 the MHRA imposed revisions to the AZD1222 Regulation 174 prescribing information to state (emphasis added), "A third dose of COVID-19 Vaccine AstraZeneca may be administered at least 8 weeks after the second dose of COVID-19 Vaccine AstraZeneca when the potential benefits outweigh any potential risk". This was considered to provide flexibility for the UK's Joint Committee on Vaccination and Immunisation to make

recommendations for all populations that may require a third dose boost of AZD1222.

Many EU NITAGs subsequently adopted recommendations for booster doses as early as 2-4 months after primary series. Some continue to recommend AZD1222 boosters at 3-4 months after primary series (Table 6 in ECDC 2022).

More recently, as detailed in Clinical Overview Section 4.3.4 regarding the Omicron variant:

> Collectively, these preliminary live virus neutralisation data suggest that 2-dose primary series immunisation with AZD1222 will likely provide limited protection against infection with the Omicron variant. These data also suggest that adding a third booster dose of AZD1222 will likely provide increased protection against infection with the Omicron variant, though still less protection than as against the original Wuhan-Hu-1 strain or other variants of concern.
>
> Despite the higher risk of breakthrough infection with Omicron due to lower nAb titres, it is considered likely that clinically meaningful protection against hospitalisation and severe infection would be maintained.

Finally, it should be noted that there were no safety issues identified with the administration of booster doses from 2.5 to 12 months after primary series vaccination.

In summary, a minimum dosing interval of 3 months would be consistent with the study design and results and would provide NITAGs with flexibility in the context of ever-evolving local conditions.

Assessment of MAH's response:

The MAH has provided the information requested.

It is now clear that a substantial proportion of the participants who contributed to the primary immunogenicity endpoint (ie, 32.7%) had a dosing interval of between 2.5 and 6 months from second dose of primary vaccination to booster dose. As detailed in Query 3, significant GMTs were observed after a booster dose of AZD1222 in subjects who received the booster < 6 months from primary series. Moreover, it appears that there is a trend of higher immunogenicity with longer interval between primary series and booster.

In conclusion, the immunogenicity results obtained from a significant proportion of subjects who received an AZD1222 booster with a dosing interval of 2.5 to 6 months from primary vaccination provides support for the claim in the SmPC that "the third dose should be administered at least 3 months after completing the primary vaccination course".

Conclusion: Point solved.

Question 13. Considering that there were differences in reactogenicity profile of AZD1222 booster dose between participants previously vaccinated with AZD1222 or mRNA vaccine by age and gender, the imbalance observed in demographic and baseline characteristics regarding age, gender and, also, the dose interval (between the booster dose and previous vaccination) between the cohorts may mask the observed reactogenicity differences between the two groups in the overall population. The MAH should explain and justify if the imbalance in demographic and baseline characteristics observed between the two groups could contribute to the difference observed in the safety profile between two cohorts.

Summary of MAH's Response:

Subgroup analyses suggest that differences in demographic and baseline characteristics may explain some, but not all, of the differences in reactogenicity between the cohorts.

Table below presents both age and sex demographics of the 4 treatment groups and the overall incidence of solicited adverse events as well as the incidences by age group (non-elderly versus elderly) and sex. (Note: differences in booster dose intervals in the 2 cohorts, with medians of approximately 4 months versus 9 months, precludes comparing reactogenicity across cohorts by dose interval.)

Consistently in all treatment groups, fewer older participants reported solicited adverse events (differences compared to non-elderly participants of between 10% and 21%) and fewer males reported solicited adverse events (differences compared to female participants of between 5% and 17%). Among the AZD1222 boosted participants, the V1222 cohort was older (median age: 62 versus 55 years) and had more male participants (54% versus 38%) than the VmRNA cohort. These imbalances could have contributed to the results seen, with the V1222 cohort's older and more male population reporting fewer events due to its age and fewer events due to its sex.

Table 47 Impact of age and sex participants reporting solicited adverse events (reactogenicity) (Seronegative safety analysis set)

	V1222/B1222	V1222/B2816	VmRNA/B1222	VmRNA/B2816
	N=347	N=349	N=299	N=301
Demographics				
Median age	62.0	63.0	55.0	55.0
% Male/Female	54/46	54/46	38/62	40/60
% Reporting solicited adverse events				
All participants	78.1	80.2	89.9	92.6
<65 years of age	87.4	88.6	95.5	95.4
≥65 years of age	67.1	70.4	74.4	85.0
Male	70.5	72.8	83.0	89.8
Female	87.1	88.8	94.1	94.4

Conversely, one can also note that the somewhat higher incidences seen in the VmRNA cohort are also seen consistently across the individual age and sex sub-groups, suggesting that difference in demographic and baseline characteristics between the cohorts were not the only reason for the somewhat higher overall reactogenicity observed in the mRNA cohort.

Reactogenicity following a first dose of AZD1222 has been consistently higher across the AZD1222 clinical development programme compared with reactogenicity following a second dose or, in study D7220C00001, reactogenicity following a third dose. Similarly, the data from study D7220C00001 suggest that reactogenicity following a booster dose of AZD1222 in those who had previously received another COVID-19 vaccine is somewhat greater than in those who previously received a 2-dose primary vaccination with AZD1222.

The conclusion that can be drawn from these data is that with more balanced cohorts of previously vaccinated AZD1222 versus mRNA participants, there may have been a smaller difference in the reactogenicity profiles of the V1222 participants versus the VmRNA participants.

Assessment of MAH's response:

An imbalance in demographic and baseline characteristic regarding age and gender between AZD1222 and mRNA cohorts was observed. Among the AZD1222 boosted participants, the cohort primed

vaccinated with AZD1222 vaccine was older (median age: 62 versus 55 years) and had more male participants (54% versus 38%) than the cohort primed vaccinated with mRNA vaccine.
In addition, in both cohort groups, fewer older participants and fewer males reported solicited adverse events. Nevertheless, the frequencies of solicited AE regarding age or gender were higher in mRNA cohort than in AZD1222 cohort.
Therefore, MAH concludes that the differences in demographic and baseline characteristics between two cohorts may explain some, but not all, of the differences in reactogenicity between them, and suggests that with more balanced cohorts of previously vaccinated AZD1222 or mRNA participants, there may have been a smaller difference in the reactogenicity profile of the AZD1222 cohort versus the mRNA cohort.

Conclusion: Point solved

Question 13. The incidence of Grade 3 solicited AEs was much higher in mRNA cohort (12.1%) than in AZD1222 cohort (1.5%), mainly due to the higher incidence of solicited grade 3 systemic AEs observed in the mRNA cohort (11.1% vs 1.2%). The severity of solicited systemic AEs after AZD1222 booster dose in mRNA cohort seemed to be higher than the primary vaccination series with AZD1222. The MAH should explain whether people previously vaccinated with mRNA who would receive a booster dose of AZD1222, could have an increased severity of reactogenicity comparing people naïve or previously vaccinated with AZD1222.

Summary of MAH's Response:

The incidence and severity of solicited AEs appears to be similar in individuals receiving an AZD1222 booster after primary series mRNA vaccine and in vaccine-naïve individuals receiving a first dose of AZD1222. Reduced incidence and severity of solicited AEs was observed following a third dose of AZD1222, consistent with information already appearing in the EU SmPC for Vaxzevria: "when compared with the first dose, adverse reactions reported after the second dose were milder and less frequent".

Table 18 compares the incidence of Grade 3 solicited AEs in the previously mRNA vaccinated participants who received an AZD1222 booster dose with incidences reported after primary AZD1222 vaccination in participants from 4 pooled COV studies. There were numerically slightly lower incidences of Grade 3 systemic solicited AEs in the naïve participants receiving a primary vaccination with AZD1222, but these differences are small, may be chance findings or due to differences in the populations (eg, the mRNA cohort was younger and more female that other cohorts, and these characteristics are associated with higher rates of reported reactogenicity), and are not considered to be clinically significant.

Table 48 Incidence of Grade 3 solicited AEs in VmRNA/B1222 participants and pooled data from naïve patients receiving AZD1222

	Study D7220C00001	Pooled Data from MAA studies
	VmRNA/B1222	AZD1222 Primary Vaccination
	n/N (%)	n/N (%)
Any Grade 3 local solicited AE	4/299 (1.3)	38/2656 (1.4)
Any Grade 3 systemic solicited AE	33/299 (11.1)	196/2664 (7.4)

The data suggest that people previously vaccinated with an mRNA-based vaccine who receive a booster dose of AZD1222 would have similar severity of reactogenicity events compared to AZD1222-naïve people and increased severity of reactogenicity events compared to people previously vaccinated with 2 doses of AZD1222.

Assessment of MAH's response:

The MAH has provided a comparative table of the incidence of severe solicited AEs after the AZD1222 booster dose in mRNA cohort and after AZD1222 primary vaccination in naïve participants from MAA pooled data.

From these data, the MAH suggests that the severity of solicited AEs after AZD1222 booster dose in subjects previously vaccinated with mRNA was similar to that in naïve participants receiving a primary vaccination with AZD1222. However, taking into account that the incidence of severe AEs was higher after the first dose of AZD1222 than after the second dose of AZD1222 during the primary vaccination course with Vaxzevria, the ideal comparison should be made between AZD1222 booster dose in mRNA and 1st dose of AZD1222 in naïve participants.

In Pooled Data from MAA studies, the incidence of severe solicited systemic AEs was 6.6% after 1st dose and 2.2% after 2nd dose of AZD1222 (similar results were reported in USA study, D8110C00001). In the other hand, the incidence of solicited grade 3 systemic AEs after AZD1222 observed in the mRNA cohort was 11.1%.

Therefore, the severity of solicited systemic AEs after AZD1222 vaccination seems to be somewhat higher in people previously vaccinated with mRNA compared to people naïve who received the 1st dose of AZD1222 (this difference is not observed in solicited local AEs).

Conclusion: Point solved.

Question 15. An assessment of the incidences of solicited AEs after the homologous or heterologous booster compared with AEs reported after primary AZD1222 vaccination should be provided.

Summary of MAH's Response:

Table 19 is a summary of solicited adverse events reported in participants in the AZD1222 booster arms in Study D7220C00001 (Seronegative Safety Analysis Set) and AZD1222 primary dose series in the Pooled Oxford Studies (Dose 1 SD for Safety Analysis Set). Overall, the proportion of participants receiving a heterologous booster with a solicited AE was generally similar with the participants receiving the primary dose series of AZD1222. Results were overall consistent for the Safety Analysis Set including seropositive participants in the AZD1222 booster arms in Study D7220C00001 (see CSR Tables 14.3.2.1.1 and 14.3.2.2.1).

Table 49 Incidence of reactogenicity events following homologous and heterologous AZD1222 booster versus primary AZD1222 vaccination

	Study D7220C00001[a]		Pooled Oxford Studies[b]
	V1222/B1222 n/N (%)	VmRNA/B1222 n/N (%)	Primary dose series of AZD1222[a] n/N (%)
Any solicited AE	**264/338 (78.1)**	**268/298 (89.9)**	**2332/2725 (85.6)**
Any local solicited AE	208/338 (61.5)	227/298 (76.2)	2002/2725 (73.5)
Tenderness	184/338 (54.4)	212/298 (71.1)	1739/2725 (63.8)
Pain	128/338 (37.9)	148/298 (49.7)	957/1762 (54.3)
Swelling	12/338 (3.6)	12/298 (4.0)	93/2704 (3.4)
Redness	10/338 (3.0)	13/298 (4.4)	84/2704 (3.1)

Bruising	NC	NC	172/963 (17.9)
Warmth	NC	NC	315/1762 (17.9)
Itch	NC	NC	356/2725 (13.1)
Induration	NC	NC	51/1762 (2.9)
Any systemic solicited AEs	**204/338 (60.4)**	**237/298 (79.5)**	**1991/2725 (73.1)**
Fatigue	142/338 (42.0)	169/298 (56.7)	1445/2725 (53.0)
Headache	114/338 (33.7)	153/298 (51.3)	1435/2725 (52.7)
Muscle pain	78/338 (23.1)	141/298 (47.3)	1197/2725 (43.9)
Malaise	72/338 (21.3)	124/298 (41.6)	783/1762 (44.4)
Chills	17/338 (5.0)	88/298 (29.5)	568/1762 (32.2)
Nausea	41/338 (12.1)	66/298 (22.1)	391/1762 (22.2)
Fever	5/338 (1.5)	30/298 (10.1)	205/2695 (7.6)
Vomiting	2/338 (0.6)	4/298 (1.3)	31/1762 (1.8)
Joint pain	NC	NC	724/2725 (26.6)
Feverishness	NC	NC	591/1762 (33.5)

Assessment of MAH's response:

The MAH provided a comparative table of the incidence of reactogenicity events following homologous and heterologous AZD1222 booster versus primary AZD1222 vaccination.
The MAH indicates that the proportion of participants receiving a heterologous booster with a solicited AE was generally similar than the proportion of participants receiving the primary dose series of AZD1222.
Additionally, according to the data from the Pooled Oxford Studies (submitted during MAA) the incidence of solicited AEs is higher after the first dose than after the second dose of AZD1222 during the primary series.
With the analysis now provided by the MAH, it is not possible to determine whether the reactogenicity after homologous or heterologous AZD1222 booster is similar or not to the known reactogenicity profile after 1st or 2nd dose of AZD1222.

Conclusion: Point solved.

Question 16. The incidence of vaccination site lymphadenopathy in the mRNA cohort was higher (frequency as "common") than the incidence included in the current SmPC as "uncommon". The MAH should discuss whether people previously vaccinated with mRNA who would receive a booster dose of AZD1222, could have an increased risk of vaccination site lymphadenopathy.

Summary of MAH's Response:

The risk of lymphadenopathy appears to be similar in those receiving a first dose of AZD1222, whether after primary series with an mRNA vaccine or in the vaccine-naïve.

The number of vaccination site lymphadenopathy adverse events reported in study D7220C0001 is provided in Table 20. A listing of the 5 patients that reported these events is provided in Table 21.

Table 50 Vaccination site lymphadenopathy adverse events reported through Day 29 (Safety analysis set)

	V1222/B1222	V1222/V2816	VmRNA/B1222	VmRNA/B2816
Preferred term	**N=367**	**N=368**	**N=322**	**N=322**
Vaccination site lymphadenopathy	0	0	3 (0.9)	2 (0.6)

The incidence for the AZD1222 boosted treatment arm is 0.93%, which thus falls under the CIOMS "uncommon" adverse drug reaction frequency (ie, reported in between <0.1% and <1% of patients). The incidence for the AZD2816 booster treatment arm is 0.62%. Due to the similarity of the 2 booster vaccinations and unlikelihood that their ADR profiles differ in this respect, it is reasonable to combine the data for a better powered estimate. Their combined incidence is 0.77%.

AstraZeneca acknowledges that lymphadenopathy is an adverse drug reaction observed following vaccination, an event which is disclosed in the current SmPC with an incidence of uncommon. Although mRNA vaccinated individuals receiving AZD1222 for the first time may be at a somewhat higher risk for these events compared to previously AZD1222 vaccinated individuals receiving their third AZD1222 dose, the incidence reported in the VmRNA cohort is in line with that reported for individuals that received a primary AZD1222 vaccination as disclosed in the current SmPC.

Assessment of the MAH's response

According to the MAH, the subjects receiving AZD1222 booster dose after mRNA primary vaccination may be at a somewhat higher risk of vaccination site lymphadenopathy compared to previously AZD1222 vaccinated individuals receiving their third AZD1222 dose.

In addition, the MAH indicates that incidence reported in the mRNA cohort is in line with that reported for individuals that received a primary AZD1222 vaccination as disclosed in the current SmPC. The assessor endorses the MAH´s position that the frequency of lymphadenopathy in mRNA cohort is ≥1/1000 and <1/100 as it has been reported in the current SmPC.

Conclusion: Point solved

Question 17. Two AEs of increased Fibrin D-dimer considered as related to AZD1222 booster dose were observed in the mRNA cohort (0.7%) compared to zero in the AZD1222 cohort. The MAH should discuss these findings and justify whether or not an increase of elevated Fibrin D-dimer should be included in the SmPC for this population group.

Summary of MAH's Response:

Coagulation safety laboratory tests, including D-dimer, were included in Day 0, Day 8, and Day 29 schedule of activities for all participants as a means of enhancing pharmacovigilance for thrombotic events. There was no increased risk for thrombotic events seen in this study, including in the few participants that reported adverse events of increased D-dimer. Overall, there were no clinically meaningful changes from baseline in D-dimer levels over time (Table 14.3.7.1.2) and results from shift tables were comparable across the cohorts (Table 14.3.7.4.2). None of the reports of elevated D-dimer in the study were serious or were associated with symptoms or a thrombotic event.

There were two increases in D-dimer in the VmRNA/B1222 that were reported as AEs and were judged by investigator to be possibly related to investigational product. No concurrent AEs were reported:

A 30-39 year old participant reported a mild severity elevated D-dimer event that was observed in Day

8 coagulation analysis: 1.56 mg/L (local lab upper limit of normal: 0.44 mg/L). No treatment was required, and the event resolved. Day 29 D-dimer: 0.25 mg/L.

A 50-59 year old participant reported a mild severity elevated D-dimer event, that was observed in Day 8 coagulation analysis: 0.67 mg/L (local lab upper limit of normal: 0.44 mg/L). No treatment was required, and the event resolved. Day 29 D-dimer: 0.27 mg/L.

The D-dimer test is considered to be highly sensitive but also non-specific. Increases in D-dimer described as small and not clinically significant have been reported following vaccination with ChAdOx1 nCoV-19 (AZD1222) (Chang et al 2022). But elevated D-dimer levels may be a result of trauma, infection, inflammation, liver or kidney disease, cancer, pregnancy, or smoking.

AstraZeneca does not consider these 2 laboratory findings, transient increases, one of which was less than 2 times the upper limit of normal and without clinical symptoms, justifies adding elevated fibrin D-dimer to the SmPC.

Assessment of the MAH's response:
A report of the two resolved events of increases in D-dimer in the mRNA cohort has been provided. The assessor endorses the MAH´s position of not adding elevated fibrin D-dimer to the SmPC

Conclusion: Point solved

Question 18. There was no imbalance in the incidence of MAAEs after AZD1222 booster dose between AZD1222 and mRNA cohorts and no new safety signal was observed. However, the MAH has not reported on the evaluation of the relationship between the MAAEs and the investigational vaccine. This analysis should be submitted.

Summary of MAH's Response:

The table below presents the number and percentage of participants reporting MAAEs that were considered possibly related to investigational product by the investigator. None of the events were serious. There were few events overall (approximately 1% per group), and no imbalance was seen.

Table 51 N (%) of participants with related medically attended adverse events – through Day 29 (Safety analysis set)

V1222/B1222 N=367	V1222/B2816 N=368	VmRNA/B1222 N=322	VmRNA/B2816 N=322
4 (1.1)	3 (0.8)	3 (0.9)	4 (1.2)

In addition, AstraZeneca reports a brief narratives of the events, presented by treatment group. These events raise no additional safety concerns

Assessment of the MAH's response:

The incidence of MAAEs reported after the booster dose was low and the frequencies similar in both groups (10.7% and 7.4% in AZD1222 and mRNA cohort, respectively).

The frequencies of MAAEs considered related to the vaccine, reported by the MAH, were very low and similar between cohorts (1.1% and 0.9% in AZD1222 and mRNA cohort, respectively).

No imbalance was observed and the narrative of the events did not raise any additional safety concern.

Conclusion: Point solved

Question 19. Regarding laboratory and haematological parameters, the MAH should provide the tables indicating the percentage and total number of participants with normal values, increased or decreased values of different laboratory and haematological parameters for groups V1222:B1222 and VmRNA:B1222

AstraZeneca's Response:

The requested shift tables are provided in Appendix B. No clinically relevant trends were observed in any treatment group

Assessment of the MAH's response:

The MAH submitted an Appendix with the coagulation and haematology and chemistry shifts from baseline to the worst severity grade on-treatment value (Through Day 29) and from baseline to the maximum/minimum on-treatment value based on normal range (Through Day 29).

The laboratory, haematological and chemistry shifts were similar between both cohorts. The majority of laboratory, haematological and chemistry parameters were within normal clinical range and did not raise any safety concerns.

Conclusion: Point solved

Question 20. Considering that trial D7220C00001 only recruited subjects > 30 yoa the MAH is asked to justify requesting a booster indication from 18 yoa. The risk of TTS in this population should also be taken into account.

Summary of MAH's Response:

The original AZD1222 development programme included participants aged 18 to 29 years. Based on the data from this programme, Vaxzevria was approved in the EU for active immunisation in individuals 18 years of age and older. In AZD1222 clinical studies, including in study D7220C00001, increased immunogenicity and increased reactogenicity have been observed in adults aged 18-64 years compared with older adults. However, clinically meaningful differences that would impact the benefit-risk profile of AZD1222 have not been observed in those aged 18 to 29 compared with, eg, those aged 30 to 39 or 40 to 49. As such, AstraZeneca considers that the booster indication should mirror the primary series indication, including with respect to minimum age.

Further, while the seronegative booster dose cohort of study D7220C00001 did not include any participants aged 18-29 years, the immunogenicity and safety of a booster dose in those aged 18-29 years is supported by the COV001 booster substudy, where of the 80 participants assessed for reactogenicity, 16 were aged 18-29, 36 were aged 30-39, and 28 were aged 40-55 (Flaxman et al 2021). No meaningful differences in immunogenicity or safety were reported across these age groups.

Thrombosis with thrombocytopenia syndrome, in some cases accompanied by bleeding, has been observed very rarely following vaccination with AZD1222. This includes severe cases presenting as venous thrombosis, including unusual sites such as cerebral venous sinus thrombosis, splanchnic vein thrombosis, as well as arterial thrombosis, concomitant with thrombocytopenia. The majority of these cases occurred within the first 21 days following vaccination and some events had a fatal outcome. The reporting rates after the second dose are lower compared to after the first dose.

There are no known risk factors for the development of thrombosis with thrombocytopenia following vaccination. Please note that in September 2021 PRAC concluded that no risk factor associated with

gender and age was identified for TTS and therefore recommended removal of 'TTS cases occurred mostly in women under 60 years of age' from section 4.4 of the SmPC.

A summary of the latest review (28 March 2022) of the AZD1222 post-marketing database regarding booster dose and potential TTS reports is provided in Appendix D. Overall, no new or emerging concern regarding TTS has been identified with booster doses of AZD1222. AstraZeneca will continue to monitor adverse event reports involving booster dosing with Vaxzevria as part of ongoing routine surveillance activities.

AstraZeneca considers that these data support the inclusion of a booster dose option in the EU SmPC in individuals 18 years of age and older. AstraZeneca is not aware of any data that would support a different age threshold for a booster dose of AZD1222 compared with primary series vaccination and considers that the overall benefit-risk profile of a booster dose is consistent with the benefit-risk profile of the primary vaccination course, regardless of age.

Assessment of the MAH's response:

In the data reported for this submission from study D7220C00001, no participants aged <30 years were included. However, the MAH considers that the safety of a booster dose in subjects aged 18-29 years is supported by the COV001 booster substudy, where from the total of 80 participants assessed for reactogenicity, 16 were aged 18-29, 36 were aged 30-39, and 28 were aged 40-55 (Flaxman et al 2021).

The assessor considers that a database of 80 participants is very limited and does not allow to draw any conclusion regarding reactogenicity and safety of the booster dose. However, it is to be expected that the reactogenicity pattern after a booster dose will be similar between subjects aged 18-29 years and subjects ≥30 years, with the exception of the frequencies profile (it is known that the reactogenicity decreases with the age). Therefore, from a point of safety view, the assessor agrees that the indication of a booster dose could include subjects from ≥18 years.

Due to the wide post-marketing use of Vaxzevria, it has been possible to characterize very rare cases of TTS following vaccination with AZD1222. In addition, the reporting rates of TTS after the second dose are lower compared to after the first dose.

The MAH attached Postmarketing Reports of Potential TTS Following AZD1222 (appendix D), but the information included is very limited. Seven cases with the HLT Thrombocytopenia and SMQ Hematopoietic were identified. Of which, there were 4 cases reported after AZD1222 booster dose: 2 of them erroneously reported to occur after dose 3, but probably appeared after dose 2; another one did not meet MHRA definition, and the last one appeared in a subject previously vaccinated with Sinovac.

With this information and without knowing the number of AZD1222 doses administrated, it is not possible to know the risk of TTS after a booster dose of AZD1222. Therefore, a warning in 4.4 in the SmPC regarding this issue should be included.

Conclusion: SmPC updated accordingly, point solved

☒Overall conclusion and impact on benefit-risk balance has/have been updated accordingly

10. Overall conclusion and impact on the benefit/risk balance

The purpose of this variation is to support the use of AZD1222 as COVID-19 vaccination booster dose in

adults 18 years and older, previously vaccinated with primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based). Consequently, an update of several sections of the SmPC is proposed by the MAH. The main supporting data derive from Study D7220C00001.

Non-clinical data

The MAH has submitted a peer reviewed publication (Spenser et al., 2021) providing information related to immunogenicity response in Balb/c mice following three doses of Vaxzevria (AZ1222). Data is indicative of an overall improved immunogenicity in this species when receiving 3 doses of the vaccine. Although no relevant increase in T-cell response has been observed, the neutralizing antibody response was overall higher after the boost dose as compared with two doses immunization against wild-type, Beta, Delta and Gamma variants in a pseudovirus nAb assay. In addition, specific IgG response against all variants assessed has shown an overall better profile as compared to two dose administration.

Of note, the MAH has submitted data with AZD2816 vaccine (modified AZD1222 vaccine targeted against the Beta variant of SARS-CoV-2). It should be highlighted that since evaluation of AZD2816 is not part of the present procedure, data related to this modified vaccine has not been considered for the assessment. In addition, information related to thromboembolic events that are currently part of other ongoing evaluation procedures of Vaxzevria (AZD1222) have been not included in the present evaluation report.

With regards to the non-clinical assessment no relevant concerns have been identified.

Clinical data

Clinical trial D7220C00001 is an ongoing Phase II/III, partially double-blinded, randomised, multinational, active controlled study to evaluate the safety and immunogenicity of AZD2816 (a modified AZD1222 vaccine targeted against the Beta variant of SARS-CoV-2) and AZD1222 (original vaccine expressing the Wuhan strain) as a 1-dose booster vaccination in previously vaccinated adult participants (either with AZD1222 or an mRNA vaccine) and also as a 2-dose primary vaccination schemes in previously unvaccinated adult participants.

It is noted however, that the MAH is not seeking an indication for the product AZD2816 at this time due to its limited relevance in an epidemiological setting dominated by Delta and Omicron variants. Moreover, data from the previously unvaccinated cohort, who are to receive a 2-dose primary series of AZD1222 and/or AZD2816, have not been submitted by the MAH in the context of this variation.

Therefore, the data submitted in this procedure by the MAH (which include an Interim CSR and a clinical overview) are primarily focused on data from the AZD1222 booster treatment (in subjects that previously received two doses of AZD1222 or an mRNA vaccine) and includes only a brief summary of results for AZD2816. It is noted that this interim analysis includes a full analysis of the booster treatment group through Day 29 following AZD1222 booster, and it is considered that these data are sufficient to get relevant information regarding the immunogenicity and safety of this booster dose.

Study participants were 30 years of age and older, and the main immunogenicity analysis is made in participants who were SARS-CoV-2 seronegative at study start.

The design of the clinical trial D7220C00001 was discussed in two Scientific Advice procedures and in one additional query about the use of an historical control group that were posed by the MAH to the CHMP. Some of the recommendations made by the CHMP in the FALs have been followed, such as the non-inferiority analysis for the GMT ratio (primary endpoint) and the seroresponse rate (key secondary endpoint), and the request to separate the original SAP into 3 individual SAPs with one specific to the AZD1222 previously vaccinated cohort, and another one to the mRNA previously vaccinated cohort. However, some CHMP recommendations were not followed by the MAH. In particular, the MAH did not

follow the CHMP recommendation on the immunogenicity comparisons to be made in order to include a claim in the product information so that AZD1222 can be used to boost the response in persons previously vaccinated with COVID-19 mRNA vaccines.

The immunogenicity comparisons were based on pseudoneutralizing antibodies and a report showing good concordance of these results with those obtained from a wild type virus neutralization assay was lacking and this issue was raised as a RSI.

It is noted that, upon consultation, the CHMP agreed that the primary and key secondary non-inferiority analyses would compare the immunogenicity, in terms of neutralizing antibodies, of the response achieved 28 days after AZD1222 booster dose as compared to that achieved in an historical control group 28 days after a 2-dose AZD1222 primary vaccine series from the Phase 3 trial, Study D8110C00001, predominantly conducted in the US and South-America. Limited details have been provided on the baseline and demographic characteristics of the historical control group, and this information was requested as a RSI in order to determine if there is a good match with the cohort that received primary vaccination with AZD1222.

The seronegative immunogenicity set used for the primary and key secondary endpoints includes 342 participants from the group that received 2 doses of AZD1222 and a homologous booster and 294 participants from the cohort that received two doses of an mRNA vaccine and a booster dose of AZD1222. The historical control group included 508 subjects.

The primary endpoint based on calculating GMT ratios between the antibody titres reached 29 days after the booster dose and those achieved 29 days after primary vaccination (derived from the historical control group) were met for both cohorts (AZD1222 and mRNA primary vaccinated subjects) whereas the key secondary endpoint comparing the seroresponse rate after primary vaccination with that pre- and post- booster dose was not met for any of the two cohorts. The low seroresponse rate achieved after the booster doses questions the indication sought for a booster AZD1222 dose, and this issue was raised as a RSI.

In conclusion, the data provided by the MAH in the initial submission of this variation were not considered sufficient to provide conclusive efficacy evidence to support de use of Vaxzevria as a booster dose, in adults 18 years and older, who were previously vaccinated with a primary series of an authorised COVID-19 vaccine (either mRNA or adenoviral-based), and this issue was raised as a major objection (MO). In particular the information requested in relation to this MO related to: i) requesting a report showing adequate correlation between the pseudovirus neutralising and live virus neutralising assays; ii, various analyses to try explaining the failure to meet the key secondary endpoint (seroresponse) and iii) justification for not following the CHMP recommendation on the immunogenicity analysis to allow the use of AZD1222 as booster for those previously vaccinated with an mRNA vaccine.

The MAH has provided adequate response to the MO and other concerns raised as RSI. As detailed in section 9. , the MAH provided histograms [showing the percentage of subjects reaching different antibody titres (pre- and post- booster)], reverse cumulative distribution curves (RCDC) of pseudoneutralising antibodies (pre- and post- booster doses), and a specific analysis of the booster responses in participants with antibody tires pre-booster below the LLOQ. Overall, the histogram data and the RCDC curves for both the V1222/B1222 and the VmRNA/B1222 cohorts do not indicate that there are two different subpopulations, one of high responders and the other responding poorly to the booster dose. Rather, the data suggest that most of the subjects that received a booster dose increased their nAb titres. For both the V1222 and VmRNA groups, at day 29 after booster doses, a higher rate of seroresponse (81.4% and 97.3%, respectively) was observed in participants with baseline nAb titres < LLOQ than in the overall population [66.1% (V1222) and 43.2% (VmRNA)]. It is thus considered that the high seroresponse rates seen in participants with baseline nAb titres < LLOQ in fact demonstrate the adequate boosting ability

of AZD1222 dose, and that the lower seroresponse rate in the overall population was due to the difficulty to achieve a ⩾ 4-fold rise in seroresponse from baseline in subjects with higher pre-booster titres.

As detailed in section 9. , the MAH also calculated the seroresponse rates based on a ⩾ 2 fold rise in Spike-binding antibodies. The seroresponse rates determined when using a ⩾ 2 fold rise in binding antibodies were 82.6% (95%CI 78- 86), and 71.1% (95%CI 65-76) for the V1222/B1222 and the VmRNA/B1222 cohorts, respectively. These figures are clearly higher from those calculated when using a ⩾ 4 fold rise in binding antibodies: 68.2% (95%C 62-73) for the V1222/B1222 and 36.7% (95%CI 30-42) for the VmRNA/B1222. These data are interpreted in that the seroresponse rate in terms of S-binding antibodies (⩾ 2 fold rise) is quite significant for both cohorts (V1222/B1222 and VmRNA/B1222), and thus these data indicate that the AZD1222 booster dose is in fact boosting the response induced after primary vaccination in most of the participants in the trial.

The MAH acknowledged that the CHMP advice has not been followed to compare the immune response of the VmRNA/B1222 group to an mRNA primary series treatment group. The MAH indicated that it was not possible to access serum samples for the VmRNA cohort nor was it possible to access mRNA vaccine for administration to a vaccine-naïve cohort within the study. Taking into account this explanation, together with: i) the fact that the comparison made by the MAH in terms of GMT ratio showed higher titers in the population that received the AZD1222 booster as compared with a population in which clinical efficacy was shown (primary series of AZD1222), ii) the high seroresponse rate seen in subjects with low antibody titres before the booster dose, and ii) the high seroconversion rate in terms of ⩾ 2 fold rise in Spike-binding antibodies, it is considered that the data submitted support the use of AZD1222 as a heterologous booster.

It is noted, that very recently a preprint publication (not yet peer-reviewed) (https://www.medrxiv.org/content/10.1101/2022.04.29.22274483v1) (Effectiveness of ChAdOx1-S COVID-19 Booster Vaccination against the Omicron and Delta variants in England) from UK, provides evidence of protection against Omicron variant following homologous AZD1222 booster.

In conclusion, all the efficacy data provided by the MAH in response to the RSI provide clear support for the use of AZD1222 as a homologous or heterologous booster.

The interim analysis of **safety data** (data cut-off 11th October 2021) was performed on 646 seronegative participants (347 participants were previously vaccinated with AZD1222 and 299 participants were previously vaccinated with a mRNA vaccine). The median number of follow-up days after AZD1222 booster dose was similar in both groups up to the cut-off date (90 days in AZD1222 cohort and 88 days in mRNA cohort). However, an imbalance regarding age, gender and the dose interval (between the booster dose and primary vaccination) was observed between the two groups. The MAH was requested to explain and justify if the imbalance in demographic and baseline characteristics observed between the two groups could contribute to the difference observed in the safety profile between the two cohorts.

Overall, the observed reactogenicity profile was similar in both groups and not different to the known reactogenicity described for AZD1222, although the frequency and severity of each solicited local and systemic AEs was higher in mRNA cohort than in AZD1222 cohort. The MAH concluded, in a response to the RSI, that the differences in demographic and baseline characteristics between two cohorts may explain some, but not all, of the differences in reactogenicity between them, and it is suggested that with more balanced cohorts of previously vaccinated AZD1222 or mRNA participants, there may have been a smaller difference in the reactogenicity profile of the AZD1222 cohort versus the mRNA cohort.

In addition, AEs were reported less frequently in adults aged ⩾ 65 years than in adults aged 30-64 years in each cohort groups and higher incidences of solicited and unsolicited AEs were observed in females than males in both cohorts. These results were consistent to data reported in previous AZD1222 studies.

No vaccine related deaths were reported during the study. The incidence of SAEs and AESIs was low and no clinically meaningful imbalances were noted. No TTS events were reported during the study.

On the other hand, there were no data from participants aged <30 years. However, it is to be expected that the reactogenicity pattern after a booster dose will be similar between subjects aged 18-29 years and subjects ⩾30 years, with the exception of the frequencies profile since it is known that the reactogenicity decreases with the age.

The benefit/risk of Vaxzevria as stated in the current PI information does not change.

In relation to this variation, the MO as well as the OCs raised as RSI have been adequately addressed by the MAH. The results indicate that both for the homologous or heterologous AZD1222 booster, the primary immunogenicity endpoint was met in both cases. Although the key secondary endpoint based on nAb seroresponse rate was not met for any of the two cohorts, the analysis described in the responses to the RSI indicate that particularly the subjects with lower vaccine-induced antibody titres before vaccination benefit clearly from a booster dose, and that a large proportion of subjects, independently of baseline titres, also increase the spike antibody titres. In conclusion, there is a clear benefit of a booster dose in terms of efficacy (increase in antibody titres).

It is noted that the data submitted do not provide information on the long-term persistence of the antibody titres.

From a safety point of view, the safety pattern is similar to that described at the time the conditional MA was granted for primary series vaccination. However, it is not possible to determine whether the reactogenicity after homologous or heterologous AZD1222 booster is similar or not to the known reactogenicity profile after 1st or 2nd dose of AZD1222. Moreover, the severity of solicited systemic AEs after AZD1222 vaccination seems to be somewhat higher in people previously vaccinated with mRNA compared to people naïve who received the 1st dose of AZD1222.

It should also be mentioned that the data submitted by the MAH do not allow to determine the risk of several rare severe adverse reactions associated with the use of AZD1222, such as capillary leak syndrome, cerebrovascular venous and sinus thrombosis, myelitis transverse and thrombosis with thrombocytopenia syndrome after AZD1222 booster dose, especially in previously mRNA-vaccinated subjects who will receive AZD1222 vaccine for the first time. The SmPC section 4.4 has been updated with a warning to indicate that the risk of very rare events after a booster dose of Vaxzevria has not been characterized.

In conclusion, the benefit/risk of a homologous booster dose of AZD1222 or a heterologous AZD1222 dose in subjects that had received an mRNA primary vaccination series is positive.

11. Recommendations

Based on the review of the submitted data, this application regarding the following change:

Variation requested		**Type**	**Annexes affected**
C.I.4	C.I.4 - Change(s) in the SPC, Labelling or PL due to new quality, preclinical, clinical or pharmacovigilance data	Type II	I and IIIB

Update of sections 4.2, 4.4, 4.8 and 5.1 of the SmPC in order to introduce a booster dose of Vaxzevria (homologous or heterologous) based on interim immunogenicity and safety data from the pivotal study D7220C00001, a partially double-blinded, randomised, multinational, active-controlled phase II/III clinical study and supportive literature evidence from studies COV001, COV-BOOST and Com-COV

studies. The Package Leaflet is updated accordingly. In addition, the MAH took the opportunity to make minor editorial changes/corrections throughout the product information.

☒is recommended for approval

Amendments to the marketing authorisation

In view of the data submitted with the variation, amendments to Annex(es) I and IIIB are recommended.

12. EPAR changes

The table in Module 8b of the EPAR will be updated as follows:

Scope

Please refer to the Recommendations section above

Summary

Please refer to Scientific Discussion 'EMEA/H/C/005675/II/0052'

For more information, please refer to the Summary of Product Characteristics.