A5404

Primary Statistical Analysis Plan

Version 3.1

January 27, 2023

SARS-CoV-2 Immune Responses after COVID-19 Therapy and Subsequent Vaccine

ClinicalTrials.gov Identifier: NCT04952402

IND # 027376

Protocol Version 2.0, CM #1

(This is ACTG A5404 SAP Version 3.1 with names of authors, names of publication writing team members and analysis timeline redacted)

Table of Contents

Version History

Version	Changes Made	Date Finalized
1	Original Version, reflecting protocol v2.0. Note that there was no enrollment under protocol v1.0.	6/9/2021
2	Updated to reflect LOA #1.	9/13/2021
3.0	Updated to reflect CM#1 which is pending final approvals; revised analysis plans due to low enrollment numbers.	17 January 2023
3.1	Minor version update to reflect that CM #1 was approved and distributed to sites on January 25, 2023.	27 January 2023

1 Introduction

1.1 Purpose

This Primary Statistical Analysis Plan (SAP) describes the primary and key secondary estimands and other secondary outcome measures that will address specific study objectives and interim monitoring of the A5404 study. The Primary SAP includes general analytic approaches for all primary estimands, key secondary estimands, and other outcome measures in the primary manuscript(s) or submitted to ClinicalTrials.gov (regardless of the reporting timeline). The Primary SAP facilitates discussion of the statistical analysis components among the lead study investigators and statisticians, helping them agree on the statistical analyses to be performed and presented in the primary analysis report.

The Analysis Implementation Plan (AIP) provides detailed outlines of tables, figures, and coding descriptions.

A separate SAP will provide outlines of analyses for other objectives and outcome measures not included in the Primary SAP.

1.2 Version History

Summary of Major Changes:

Version 1.0: Not applicable; original version.

Version 2.0: Modifications per LOA #1

- Modifications to the treatment regimens throughout to match revised language in LOA #1.
- Modifications to general language throughout to match revised language in LOA #1.
- Revision to Section 2.5 to reflect updated trigger for first SMC review per LOA #1.
- Modifications throughout to reflect revised time points for outcome measures 3.1.1, 3.2.2, and 3.2.6.
- Modification to Retrospective Safety Set to allow for inclusion of participants who received both doses of vaccine prior to study entry.

Version 3.0: Modifications in anticipation of reduced sample size for the final analysis and modifications to align with CM #1.

- Revised naming of mRNA vaccination regimens (to align with CM #1)
- Modifications to text to clarify initially intended analyses and introduce revised plans in the event of reduced final sample size
    - Primary analyses will no longer focus within each select therapy group. Active therapies will be combined together into a single group.
    - Remove references to “active comparator” group since no participants enrolled from an active comparator in ACTIV-2/A5401

  - Focus will be on descriptive analyses among participants who received active therapies in ACTIV-2/A5401, those who received placebo, those who received Camostat or placebo.
  - Revised language to state that formal comparisons between groups may not be justified due to small sample size.
- Removal of Retrospective Safety Analysis Set (no longer relevant).
- Revisions to Report Contents corresponding to changes throughout.
- Revisions to remove secondary objectives and corresponding outcome measures according to CM #1.

NOTE: SAP version 3.0 aligns with protocol version 2.0 plus CM #1. At the time of this SAP version finalization (January 17, 2023), CM #1 has not yet been released to sites for implementation. The A5404 protocol v2.0 was under revision since September 2021. The team have faced a number of decisions that changed the course of action on multiple occasions for how best to proceed with changes to the protocol. The SAP was updated (in draft state) concurrent with all protocol revisions. The final analysis report for A5404 was conducted under this SAP version 3.0. The analyses align with protocol version 2.0/CM #1.

Version 3.1: Minor revision to indicate that CM #1 was distributed to sites on January 25, 2023.

2 Study Overview

2.1 Study Design

A5404 is a phase IV, open-label, sub-study of ACTIV-2/A5401 aiming to evaluate how prior investigational therapy for COVID-19 versus comparator (placebo or active comparator) affects vaccine response. This is a non-randomized study designed to have reasonable precision to estimate the difference in immune response between those with versus without prior select investigational therapy for COVID-19. Results will be interpreted in the context of available information from outside the study when analyses are completed.

The study will enroll a target of 70 participants from each ACTIV-2/A5401 select therapy group (comprised of individuals exposed to active treatment and individuals exposed to the corresponding placebo or active comparator). In addition, up to 70 individuals without prior history of SARS-CoV-2 infection will be enrolled based on vaccine supply at each A5404 site. The total sample size will depend on how many therapies are selected from ACTIV-2/A5401. The ratio of participants with active therapy exposure versus corresponding comparator exposure in ACTIV-2/A5401 is also unknown a priori, as the randomization ratio for ACTIV-2/A5401 study depends on the number of agents concurrently enrolling. If the target sample of 70 participants for a select therapy group is not able to be met from Phase II participants in the ACTIV-2/A5401 trial, then enrollment will open to participants from the respective Phase III group in ACTIV-2/A5401.

Eligible participants will receive mRNA COVID-19 vaccine in one of three ways:

Cohort 1: ACTIV-2/A5401 participants

Participants will receive one of the following regimens:

Cohort 1a: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered intramuscularly (IM) at Entry and Day 28. Both doses will be provided by the study.

Or

Cohort 1b: Participants will receive a two-dose series of a community-provided mRNA-based COVID-19 vaccine that has received FDA EUA or FDA approval (e.g., Moderna or Pfizer). Vaccine will not be provided by the study.

NOTE: These participants may enter the study before their first vaccine dose, after their first vaccine dose, or after their second vaccine dose.

Or

Cohort 1c: Participants will receive their first Moderna mRNA-1273 COVID-19 vaccine dose in the community. Their second Moderna mRNA-1273 dose will be provided by the study (100 µg [0.5 mL] to be administered IM 28 days after their first dose).

NOTE: These participants may enter the study before or after their first vaccine dose.

Or

Cohort 2: Participants without prior history of SARS-CoV-2 infection

Participants will receive the following study-provided regimen: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered IM at Entry and Day 28.

All participants will have blood collected and immune responses measured before the first vaccine dose (if feasible), and before the second dose of vaccine (e.g., 28 days later for Moderna vaccine and 21 days later for Pfizer vaccine if given in the community) (if feasible), and 56 (if feasible), 140, 365, and 730 days after their first vaccine dose.

We initially intended to conduct the primary analysis separately within each select ACTIV-2/A5401 therapy group. However, due to a lower than anticipated accrual, we revised the plan to instead conduct the primary analysis combining participants from select ACTIV-2/A5401 therapy groups. The primary analysis will occur after the last enrolled participant from ACTIV-2/A5401 has reached the Day 140 study visit.

2.2 Hypotheses

This study hypothesizes that there may be differences in immune response following mRNA-based vaccination for SARS-CoV-2 among individuals with prior SARS-CoV-2 infection between those exposed to prior select therapies versus those who received a placebo. This study focuses on descriptive objectives rather than formal statistical hypothesis testing.

2.3 Study Objectives

This Primary SAP addresses the following primary and secondary objectives listed in the study protocol. Other study objectives in the protocol will be addressed in subsequent analysis plans.

The primary and secondary study objectives below will be analyzed descriptively and conducted only if sample size permits. The primary objective analysis will be conducted once the last enrolled participant for that group has completed the Day 140 study visit and all associated queries have been resolved. Analysis of the secondary objective will occur at the same time.

Note: In CM #1, four of the originally stated secondary objectives were relegated to exploratory/other objectives due to low enrollment.

2.3.1 Primary Objective

To estimate the difference in neutralizing antibody (NAb) response to mRNA-based COVID-19 vaccine among participants with prior SARS-CoV-2 infection who received investigational therapy exposure compared to participants without prior investigational therapy exposure (placebo for investigational therapy or active comparator).

2.3.2 Secondary Objective

To explore the safety of mRNA-based COVID-19 vaccines among participants with prior COVID-19 who received investigational therapy or its comparator for COVID-19 and persons who are SARS-CoV-2 naïve.

2.4 Overview of Sample Size Considerations

The study is descriptive and designed to have reasonable precision around the primary outcome measure estimate. See Section 10.4 of the study protocol (version 2.0) and CM #1 for additional details. In January 2022, the study team noted that the observed enrollment into the study was lower than anticipated and that achieving the target sample size of 70 participants per select therapy group was unlikely. The study closed to accrual on February 25, 2022, and enrolled a total of 43 participants with no enrollment of participants exposed to active comparator in ACTIV-2/A5401. Therefore, we have revised the analyses as necessary and stated alternatives for use with small sample sizes.

For the first primary outcome measure (NAb levels 140 days after the first vaccine dose, see SAP Section 3.1.1), we will estimate the NAb levels 140 days after the first vaccine dose in each of the following groups:

(1) Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401
(2) Participants exposed to placebo in ACTIV-2/A5401
(3) Participants exposed to placebo or Camostat Oral in ACTIV-2/A5401

We will also estimate the ratio of geometric mean responses (GMRs) for participants with COVID-19 therapy exposure (group 1 above) versus participants who received placebo (group 2 above). The precision on this estimate may be low due to limited sample size. A ratio of GMRs (GMR for select therapy exposed divided by GMR for placebo) of 1 indicates no difference in NAb response. A ratio less than 1 indicates lower NAb response among those who received COVID-

19 investigational therapy exposure compared to those who received placebo. In a sensitivity analysis we will consider a comparison of group 1 versus group 3.

A sample size of 70 participants gives reasonable anticipated precision for the ratio of GMRs. For a standard deviation of 0.5 and a sample size of 70 participants who receive vaccine (a 1:1 ratio; 35 with prior select therapy and 35 who received placebo), the 90% confidence interval for the ratio of GMRs is (0.63, 1.58) and the 95% confidence interval is (0.58, 1.73). These confidence intervals suggest reasonable precision to rule out reductions in response associated with prior select therapy of about 40% or more, or increases of about 60% or more. However, with sample sizes of 20 or 30, we anticipate wide confidence intervals (low precision) and therefore limited interpretation.

The anticipated precision for the estimate of the key secondary outcome measure, the relative change in NAb level pre-vaccine to post-vaccine (see SAP Section 3.2.1), is also reasonable for a sample size of 70. Assuming a variability of changes in NAb level of SD=0.4, a sample size of 70 participants receiving vaccine with equal numbers with versus without prior investigational therapy, the 90% confidence interval is (0.69, 1.44) and the 95% confidence interval is (0.64, 1.55) when the observed ratio of GMRs comparing participants with versus without prior select therapy is 1. However, with sample sizes of 20 or 30, we anticipate wide confidence intervals (low precision) and therefore limited interpretation.

2.5 Overview of Formal Interim Monitoring

The study will undergo safety reviews by an ACTG-appointed Study Monitoring Committee (SMC). The first interim review will occur when 10 participants have enrolled to one select therapy group, received their first dose of vaccine after study entry, and their data through study Day 7 (7 days after the first vaccine dose) are available, or 3 months after the first participant enrolls, whichever occurs first. Subsequent reviews will occur every 6 months unless otherwise recommended by the SMC or requested by the study team.

It is not intended that the SMC will review data on immunologic outcomes. An interim review may also be convened if a safety concern is identified by the DAIDS clinical representative, the study chairs, or study statistician in consultation with the team. Enrollment will pause and the SMC will review any death that occurs on study that is deemed related to study product as determined by the site investigator. A pause in enrollment will also occur and the SMC will review if two participants experience a Grade 4 adverse event (AE) that is deemed related to study product as determined by the site investigator.

3 Outcome Measures

3.1 Primary Outcome Measures

3.1.1 Neutralizing antibody (NAb) level at least 140 days after the first dose of the study- or community-provided vaccine. (Objective 2.3.1.1)

NAb level will be measured using the pseudotyped virus reported from a single-round-of-infection neutralization assay (PSV).

3.2 Secondary Outcome Measures

3.2.1 Relative pre-vaccine to post-vaccine change in NAb response defined as the ratio of post-vaccine level/pre-vaccine level. The pre-vaccine NAb measurement will be obtained before the first dose of the vaccine and the post-vaccine measurement will be obtained at least 56 days after the first dose of the vaccine. (Objective 2.3.1.1)

3.2.2 New Grade 3 or higher AE, or SAE, or AE leading to change or discontinuation in vaccine receipt from first dose of the mRNA-based COVID-19 vaccine and through 140 days after the first dose of vaccine. (Objective 2.3.2.1)

3.2.3 Grade 1 or higher allergic reaction from first dose of the mRNA-based COVID-19 vaccine through the visit 56 days after the first dose of the vaccine. (Objective 2.3.2.1)

3.2.4 Grade 2 or higher injection site reaction from first dose of the mRNA-based COVID-19 vaccine through the visit 56 days after the first dose of the vaccine. (Objective 2.3.2.1)

3.2.5 Relative pre-vaccine to post-vaccine change in NAb response defined as the ratio of post-vaccine level/pre-vaccine level by received vaccine, i.e., Moderna versus Pfizer. The pre-vaccine NAb measurement will be obtained before the first dose of the vaccine, and the post-vaccine measurement will be obtained at least 56 days after the first dose of the vaccine. (Objective 2.3.1.1)

4 General Considerations

The aim of this study is to describe immunologic responses to mRNA-based SARS-CoV-2 vaccine among individuals who previously received investigational therapy in ACTIV-2/A5401 for SARS-CoV-2 infection in comparison to individuals who received a placebo. As ACTIV-2/A5401 is expected to change from using a placebo control to an active control, this study's aim will also evolve to describe immunologic responses to mRNA-based SARS-CoV-2 vaccine among individuals who previously received investigational therapy in ACTIV-2/A5401 for SARS-CoV-2 infection in comparison to individuals who received an active control.

Estimates and confidence intervals for NAb will be obtained by using a log base 10 transformation on the PSV values, then using linear regression with an indicator variable for prior select COVID-19 therapy exposure versus placebo using standard methods, assuming that the $\log_{10}$ NAb responses are approximately normally distributed within exposure group.

4.1 Analysis Sets and subgroups

4.1.1 ACTIV-2/A5401 active therapy/placebo Response Set

All enrolled participants who were previously enrolled to active therapy or placebo in ACTIV-2/A5401 who received at least one dose of vaccine.

4.1.2 Safety Set

All enrolled participants who received the first dose of vaccine

Subgroups or subpopulations of interest

4.1.3 Subgroups defined by levels of sex, if sample size permits

4.1.4 Subgroups defined by levels of race/ethnicity, if sample size permits

5 Estimands

5.1 Primary Estimand

Primary Objective 2.3.1.1: To estimate the difference in neutralizing antibody (NAb) response to mRNA-based COVID-19 vaccine among participants with prior SARS-CoV-2 infection who received investigational therapy exposure compared to participants without prior investigational therapy exposure (placebo for investigational therapy).	
Estimand description	Neutralizing antibody response measured at least 140 days after the first dose of the study- or community-provided mRNA-based COVID-19 vaccine in individuals who previously received therapy for COVID-19 (active therapy versus placebo) and who received at least one dose of an mRNA-based COVID-19 vaccine.
Treatment	Cohort 1a: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered intramuscularly (IM) at Entry and Day 28. Both doses will be provided by the study. Or Cohort 1b: Participants will receive a two-dose series of a community-provided mRNA-based COVID-19 vaccine that has received FDA EUA or FDA approval (e.g., Moderna or Pfizer). Vaccine will not be provided by the study. Or Cohort 1c: Participants will receive their first Moderna mRNA-1273 COVID-19 vaccine dose in the community. Their second Moderna mRNA-1273 dose will be provided by the study (100 µg [0.5 mL] to be administered IM 28 days after their first dose).

Target population	Analysis sets
Individuals who received an mRNA-based COVID-19 vaccine who previously received therapy for COVID-19 infection.	ACTIV-2/A5401 active therapy/placebo Response set (All enrolled participants who were previously enrolled to active therapy or placebo in ACTIV-2/A5401 who received at least one dose of vaccine.)
Variables	**Outcome measure**
NAb level measured approximately 140 days after receiving the first dose of an mRNA-based vaccine for COVID-19.	Neutralizing antibody (NAb) level at least 140 days after the first vaccine dose.
Handling of intercurrent events	**Handling of missing data**
The following intercurrent events are relevant to the estimand: 1. Reinfection with SARS-CoV-2 between first and second vaccine doses. *Post-vaccination observations before reinfection will be used to determine the variable (while alive and prior to reinfection strategy)* 2. Reinfection with SARS-CoV-2 after second vaccine dose. *Post vaccination observations before reinfection will be used to determine the variable (while alive and prior to reinfection strategy)* 3. Discontinuation of treatment after first vaccine dose due to safety event(s).	If the day 140 outcome is missing, we will use the value from day 56 (last observation carried forward method of imputation). Participants missing both day 56 and day 140 study visit outcome data will be excluded from the analysis. Sensitivity Analyses: 1. We will impute the missing Day 140 outcome values using multiple imputation (if sufficient sample size) 2. We will only include participants who provided a Day 140 NAb measurement.

Post vaccination observations before discontinuation will be used to determine the variable (while alive and prior to discontinuation strategy) 4. Discontinuation of treatment after first vaccine dose due to non-safety reasons. *Post vaccination observations before discontinuation will be used to determine the variable (while alive and prior to discontinuation strategy)* 5. Death prohibiting subsequent existence of the variable. *Observations before death will be used to determine the variable (while alive strategy)*	
Population-level summary measure	**Analysis approach**
Geometric mean neutralizing antibody levels at 140 days after first dose of an mRNA-based COVID-19 vaccine.	We will report the geometric mean NAb levels among each group: (1) Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401 (2) Participants exposed to placebo in ACTIV-2/A5401 (3) Participants exposed to placebo or Camostat Oral in ACTIV-2/A5401 Ratio of geometric mean neutralizing antibody levels comparing those who received select investigational therapy (AZD7442 IM or IV, BRII-196+BRII-198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401) relative to placebo at Day 140 study visit, together with corresponding 95% confidence interval. We will use the log base 10 scale for the analysis of NAb measurements and report the anti-logged estimates and confidence intervals. We assume that the NAb responses are approximately normally distributed on the log base 10 scale and so confidence intervals can be calculated using standard methods. The analysis will be repeated comparing those who received select investigational therapy (AZD7442 IM or IV, BRII-196+BRII-198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401) relative to placebo or Camostat.

Supplemental Analyses

To allow for the possibility that the distribution of time from receipt of select investigational therapy or placebo to the first dose of vaccine may differ, an analysis adjusted for this time may also be undertaken if sample size permits.

If sample size permits, the same analyses will be carried out in subgroups defined by levels of sex, race/ethnicity, and mRNA vaccine (Moderna or Pfizer).

5.2 Key Secondary Estimand

Primary Objective 2.3.1.1: To estimate the difference in neutralizing antibody (NAb) response to mRNA-based COVID-19 vaccine among participants with prior COVID-19 who received investigational therapy exposure compared to participants without prior investigational therapy exposure (placebo for the investigational therapy).	
Estimand description	Neutralizing antibody response measured before and at least 56 days after the first dose of the study- or community-provided mRNA-based COVID-19 vaccine in individuals who received at least one dose of an mRNA-based COVID-19 vaccine.
Treatment	Cohort 1a: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered intramuscularly (IM) at Entry and Day 28. Both doses will be provided by the study. Or Cohort 1b: Participants will receive a two-dose series of a community-provided mRNA-based COVID-19 vaccine that has received FDA EUA or FDA approval (e.g., Moderna or Pfizer). Vaccine will not be provided by the study. Or Cohort 1c: Participants will receive their first Moderna mRNA-1273 COVID-19 vaccine dose in the community. Their second Moderna mRNA-1273 dose will be provided by the study (100 µg [0.5 mL] to be administered IM 28 days after their first dose).

Target population	Analysis set
Individuals who received an mRNA-based COVID-19 vaccine who previously received therapy for COVID-19 infection.	All enrolled participants who were previously enrolled to active therapy or placebo in ACTIV-2/A5401 who received at least one dose of vaccine and have a NAb level from before first vaccine dose and a NAb level from at least 56 days after the first dose of the vaccine.
Variables	**Outcome measure**
NAb level measured prior to receiving the first dose of an mRNA-based vaccine for COVID-19. NAb level measured approximately 56 days after receiving the first dose of an mRNA-based vaccine for COVID-19.	Relative pre-vaccine to post-vaccine change in NAb response defined as the ratio of post-vaccine level/pre-vaccine level. The pre-vaccine NAb measurement will be obtained before the first dose of the vaccine and the post-vaccine measurement will be obtained at least 56 days after the first dose of the vaccine.
Handling of intercurrent events	**Handling of missing data**
The following intercurrent events are relevant to the estimand: 1. Reinfection with SARS-CoV-2 between first and second vaccine doses. *Post vaccination observations before reinfection will be used to determine the variable (while alive and prior to reinfection strategy)* 2. Reinfection with SARS-CoV-2 after second vaccine dose. *Post vaccination observations before reinfection will be used to determine the variable (while alive and prior to reinfection strategy)*	For participants who receive the first dose of vaccine prior to study entry, the pre-first vaccine dose NAb measurement will be missing and these individuals will be excluded from the analysis (per analysis set defined above). If the day 56 study visit outcome is missing, we will use the value from the day 28 study visit (last observation carried forward method of imputation). Participants missing both day 28 and day 56 study visit outcome data will be excluded from the analysis.

3. Discontinuation of treatment after first vaccine dose due to safety event(s). *Post vaccination observations before discontinuation will be used to determine the variable (while alive and prior to discontinuation strategy)* 4. Discontinuation of treatment after first vaccine dose due to non-safety reasons. *Post vaccination observations before discontinuation will be used to determine the variable (while alive and prior to discontinuation strategy)* 5. Death prohibiting subsequent existence of the variable. *Observations before death will be used to determine the variable (while alive strategy)*	Sensitivity Analyses: 1. We will impute the missing outcome values 56 days after the first vaccine dose using multiple imputation. 2. We will only include participants who had NAb measurements at Entry and 56 days after the first vaccine dose.
Population-level summary measure	**Analysis approach**
Relative change in geometric mean neutralizing antibody levels from before first vaccine dose to 56 days after first vaccine dose.	We will report the relative change in geometric mean NAb levels among each group: (1) Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401 (2) Participants exposed to placebo in ACTIV-2/A5401 (3) Participants exposed to placebo or Camostat Oral in ACTIV-2/A5401 Ratio of change in geometric mean neutralizing antibody levels comparing those who received select investigational therapy (AZD7442 IM or IV, BRII-196+BRII-198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401) relative to placebo from before first vaccine dose to Day 56 study visit, together with corresponding 95% confidence interval. We will use the log base 10 scale for the analysis of NAb measurements and report the anti-logged estimates and confidence intervals. We assume that the NAb responses are approximately normally distributed on the log base 10 scale and so confidence intervals can be calculated using standard methods. The analysis will be repeated comparing those who received select investigational therapy (AZD7442 IM or IV, BRII-196+BRII-198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401) relative to placebo or Camostat.

Supplemental Analyses

To allow for the possibility that the distribution of time from receipt of select investigational therapy or placebo to the first dose of vaccine may differ, an analysis adjusted for this time may also be undertaken if sample size permits.

If sample size permits, analyses will be carried out in subgroups defined by levels of sex and race/ethnicity.

5.3 Secondary Safety Estimand

Secondary Objective 2.3.2.1: To explore the safety of mRNA-based COVID-19 vaccines among participants with prior COVID-19 who received investigational therapy or its comparator for COVID-19 and persons who are SARS-CoV-2 naïve.	
Estimand description	Occurrence of adverse events up to 56 and 140 days after receiving the first dose of an mRNA-based COVID-19 vaccine among individuals receiving at least one dose of the vaccine with and without prior history of COVID-19.
Treatment	Cohort 1a: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered intramuscularly (IM) at Entry and Day 28. Both doses will be provided by the study. Or Cohort 1b: Participants will receive a two-dose series of a community-provided mRNA-based COVID-19 vaccine that has received FDA EUA or FDA approval (e.g., Moderna or Pfizer). Vaccine will not be provided by the study. NOTE: These participants may enter the study before their first vaccine dose, after their first vaccine dose, or after their second vaccine dose. Or Cohort 1c: Participants will receive their first Moderna mRNA-1273 COVID-19 vaccine dose in the community. Their second Moderna mRNA-1273 dose will be provided by the study (100 µg [0.5 mL] to be administered IM 28 days after their first dose). Or Cohort 2: Participants will receive the following study-provided regimen: Moderna mRNA-1273 COVID-19 vaccine, 100 µg (0.5 mL) to be administered IM at Entry and Day 28.

Target population	Analysis sets
Individuals who received an mRNA-based COVID-19 vaccine with and without a prior history of COVID-19.	Safety Set (all enrolled participants who received at least one dose of vaccine)
Variable	**Outcome measures**
Occurrence of an adverse event up to 56 and 140 days after first dose of an mRNA-based COVID-19 vaccine.	New Grade 3 or higher AE, SAE, or AE leading to change or discontinuation in vaccine receipt from first dose of the mRNA-based COVID-19 vaccine and through 140 days after the first dose of the vaccine. Grade 1 or higher allergic reaction from first dose of the mRNA-based COVID-19 vaccine through the visit 56 days after the first dose of the vaccine. Grade 2 or higher injection site reaction from first dose of the mRNA-based COVID-19 vaccine through the visit 56 days after the first dose of the vaccine.
Handling of intercurrent events	Handling of missing data

The following intercurrent events are relevant to the estimand: 1. Reinfection with SARS-CoV-2 between first and second vaccine doses. *Post vaccination observations through the respective time point of interest (56 or 140 days) after first vaccine dose will be used to determine the variable (treatment policy strategy)* 2. Discontinuation of treatment after first vaccine dose due to safety event(s). *All observations through the respective time point of interest (56 or 140 days) after first vaccine dose will be used to determine the variable (treatment policy strategy)* 3. Discontinuation of treatment after first dose of vaccine due to non-safety reasons. *All observations through the respective time point of interest (56 or 140 days) after first vaccine dose will be used to determine the variable (treatment policy strategy)* 4. Death prohibiting subsequent existence of the variable. *Observations before death will be used to determine the variable (while alive strategy)*	Participants who discontinue follow-up before the respective time point of interest (56 or 140 days) after the first dose of the vaccine will have their outcome measures determined based on data available until the time of study discontinuation. This implicitly assumes that the participants would not have had an applicable AE had they been observed to the time point of interest. Sensitivity analyses: We will consider those participants who discontinued follow-up before the time point of interest (56 or 140 days) after the first vaccine dose as having at least one adverse event. We will only include participants who were not re-infected with SARS-CoV-2 between first and second doses of the vaccine.
Population-level summary measure	**Analysis approach**
Probability of having at least one Grade 3 or higher adverse event following first dose of vaccine through 140 days after the first vaccine dose. Probability of having a Grade 1 or higher allergic reaction from the first dose of vaccine through 56 days after the first vaccine dose. Probability of having a Grade 2 or higher injection site reaction from first dose of vaccine through 56 days after the first vaccine dose.	Proportions and confidence intervals described below will be estimated for: (1) Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous or Camostat in ACTIV-2/A5401 (2) Participants exposed to placebo in ACTIV-2/A5401 (3) Participants without prior history of COVID-19 (note that for the safety analysis, Camostat is considered an active agent) Proportion of participants with at least one Grade 3 or higher adverse event through 140 days after the first vaccine dose and exact 95% confidence interval. Proportion of participants with a Grade 1 or higher allergic reaction from the first dose of vaccine through 56 days after the first vaccine dose and exact 95% confidence interval. Proportion of participants with a Grade 2 or higher injection site reaction from first dose of

	vaccine through 56 days after the first vaccine dose and exact 95% confidence interval.

Supplemental Analyses

If sample size permits, analyses will be carried out in subgroups defined by levels of sex and levels of race/ethnicity.

6 Analysis of other secondary objectives

6.1.1 Relative pre-vaccine to post-vaccine change in NAb response defined as the ratio of post-vaccine level/pre-vaccine level by received vaccine, i.e., Moderna versus Pfizer. The pre-vaccine NAb measurement will be obtained before the first dose of the vaccine, and the post-vaccine measurement will be obtained at least 56 days after the first dose of the vaccine. (Objective 2.3.1.1)

This outcome measure will follow the Key Secondary Estimand analysis outlined in Section 5.2. We will report the relative change in geometric mean NAb levels among those who received the Moderna vaccine versus the Pfizer vaccine. We will report the ratio of change in geometric mean neutralizing antibody levels comparing those who received Moderna to Pfizer from before first vaccine dose to Day 56 study visit, together with the corresponding 95% confidence interval.

7 Report Contents

All tables, listings, and figures provided in the final report will include totals and summary statistics overall and by select investigational therapy group from ACTIV-2/A5401 (by receipt of active therapy, placebo, or among the 3 groups listed in Section 2.4).

7.1 CONSORT Diagram

7.2 Study Entry

- Accrual by site, select therapy group, and month
- Summary of eligibility violations

7.3 Baseline Characteristics

- Summary of age, sex, gender, race, ethnicity, and BMI
- Summary of baseline NAb level

7.4 Study Status

- Summary of off-study reasons
- Summary of number of days on study

7.5 Treatment Status

- Summary of number of participants who discontinued treatment after first dose of vaccine
- Summary of number of participants who received both doses of vaccine
- Summary of number of participants who did not receive first dose of vaccine (late exclusions)
- Summary of number of participants who received Moderna, Pfizer vaccine
- Summary of number of participants who received vaccine prior to study entry
    - Summary of days from first vaccine to study entry
- Summary of number of participants who received:
    - both doses on study
    - both doses in the community
    - first dose in the community and second dose on study.
- For participants from A5401/ACTIV-2
    - summary of days from onset of signs/symptoms to first A5401 treatment
    - summary of days from last A5401 treatment to first dose of vaccine

7.6 Adverse Events

- Summary of post-entry reportable adverse events (per section 7.0 of study protocol)
- Listing of all adverse events by participant (public participant identifier)
- Summary of post-entry grade 3 or higher AE, SAE, or AE leading to a change or discontinuation in vaccine receipt from first dose of the mRNA-based COVID-19 vaccine through 140 days after the first vaccine dose

- Summary of post-entry grade 1 or higher allergic reactions from first dose of the mRNA-based COVID-19 vaccine through 56 days after the first vaccine dose
- Summary of post-entry grade 2 or higher infection site reactions from first dose of the mRNA-based COVID-19 vaccine through 56 days after the first vaccine dose

7.7 Primary Estimand

- Geometric mean neutralizing antibody levels (both ND50 and ND80 titers) at 140 days after the first vaccine dose among
    - Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401
    - Participants exposed to placebo in ACTIV-2/A5401
    - Participants exposed to placebo or Camostat in ACTIV-2/A5401
- Ratio of geometric mean neutralizing antibody levels (comparing those who received select therapy relative to placebo) at 140 days after the first vaccine dose, together with corresponding 95% confidence interval.
- Ratio of geometric mean neutralizing antibody levels (comparing those who received select therapy relative to placebo or Camostat) at 140 days after the first vaccine dose, together with corresponding 95% confidence interval.
- Sensitivity analyses using multiple imputation for those with missing NAb measurement 140 days after the first vaccine dose (if sufficient sample size).
- Sensitivity analyses conducted only among participants who had Day 140 study visit NAb measurement available.
- Supplemental analyses adjusting for time from receipt of select investigational ACTIV-2/A5401 therapy (active or placebo) to the first dose of vaccine (if sufficient sample size).
- Supplemental analyses among levels of sex, race/ethnicity, and mRNA vaccine (Moderna or Pfizer) (if sufficient sample size).

7.8 Key Secondary Estimand

- Change in geometric mean neutralizing antibody levels from before first vaccine dose to 56 days after the first vaccine dose among
    - Participants exposed to AZD7442 IM or IV, BRII-196+BRII 198 IV, SAB 185 (3,840 or 10,240 units/kg), BMS 096414+BMS 986413 subcutaneous in ACTIV-2/A5401
    - Participants exposed to placebo in ACTIV-2/A5401
    - Participants exposed to placebo or Camostat Oral in ACTIV-2/A5401
- Ratio of change in geometric mean neutralizing antibody levels (comparing those who received select therapy relative to placebo) from before first vaccine dose to 56 days after the first vaccine dose, together with corresponding 95% confidence interval.
- Ratio of change in geometric mean neutralizing antibody levels (comparing those who received select therapy relative to placebo or Camostat) from before first vaccine dose to 56 days after the first vaccine dose, together with corresponding 95% confidence interval.

- Sensitivity analyses using multiple imputation for those with missing NAb measurement 56 days after the first vaccine dose (if sufficient sample size).
- Sensitivity analyses conducted only among participants who had NAb measurements at Entry and 56 days after the first vaccine dose.
- Supplemental analyses adjusting for time from receipt of select investigational ACTIV-2/A5401 therapy (active or placebo) to the first dose of vaccine (if sufficient sample size).
- Supplemental analyses among levels of sex and race/ethnicity(if sufficient sample size).

7.9 Secondary Safety Estimand

- Probability of having at least one Grade 3 or higher adverse event through 140 days after the first vaccine dose.
- Probability of having a Grade 1 or higher allergic reaction from the first dose of vaccine through 56 days after the first vaccine dose.
- Probability of having a Grade 2 or higher injection site reaction from first dose of vaccine through 56 days after the first vaccine dose.
- Sensitivity analysis considering those participants who discontinued follow-up before 56 days/140 days (respective to each outcome measure) after the first vaccine dose as having at least one adverse event.
- Sensitivity analysis only among participants who were not re-infected with SARS-CoV-2 between first and second doses of the vaccine.

7.10 Secondary Outcome Measures

- Ratio of change in geometric mean neutralizing antibody levels (comparing those who received Moderna to Pfizer vaccine) from before first vaccine dose to 56 days after the first vaccine dose, together with corresponding 95% confidence interval.