October 2, 2023

Cooperative Agreement IP-22-004: US Platform to Measure Effectiveness of Seasonal Influenza, COVID-19 and other Respiratory Virus Vaccines for the Prevention of Acute Illness in Ambulatory Settings

Component D: Immunologic Study of Response to Influenza and COVID-19 Virus Vaccination

Study Title: Randomized study of the immunogenicity and duration of antibody response against circulating SARS-CoV-2 variant and influenza viruses following concomitant versus sequential administration of mRNA COVID-19 vaccine and quadrivalent cell culture-based influenza vaccine among children and adults

Short Title: Comparative immunogenicity of concomitant vs sequential mRNA COVID-19 and influenza vaccinations

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Version Number: 2

2 October 2023

STATEMENT OF COMPLIANCE

- This trial will be conducted in compliance with the protocol, the International Conference on Harmonization (ICH) Guideline E6—Good Clinical Practice (GCP), and the applicable guidelines and regulatory requirements from the United States (US) Code of Federal Regulations (CFR), 45 CFR Part 46.
- All study personnel with participant contact have completed Human Participants Protection Training.

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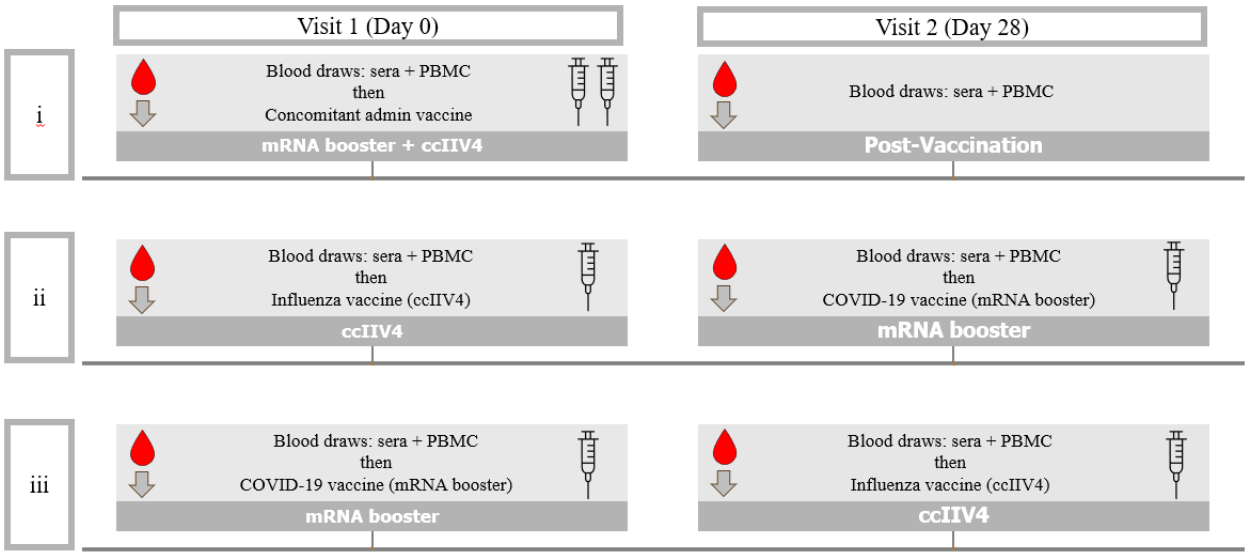
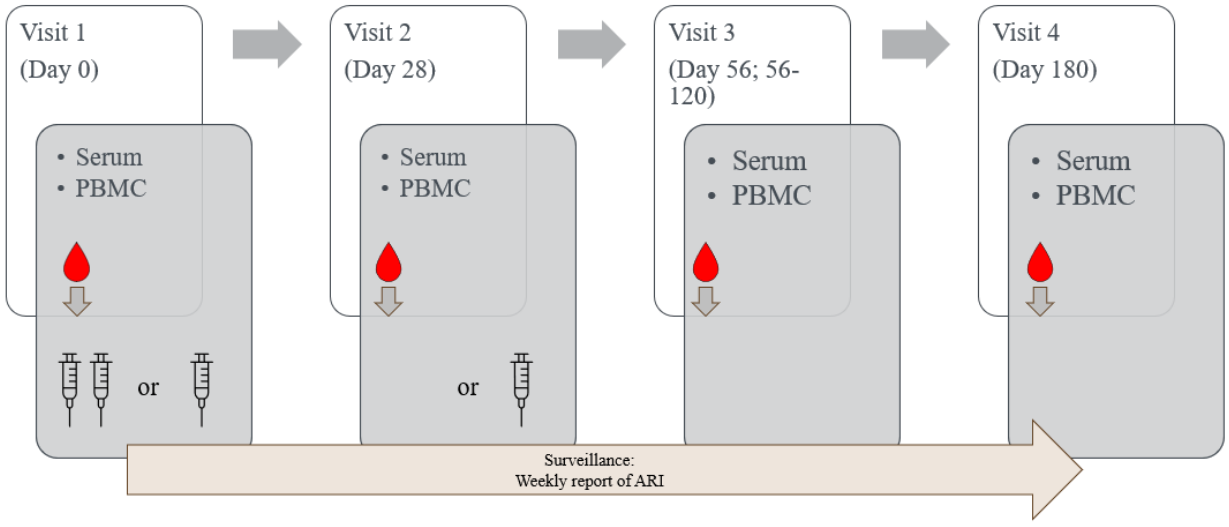
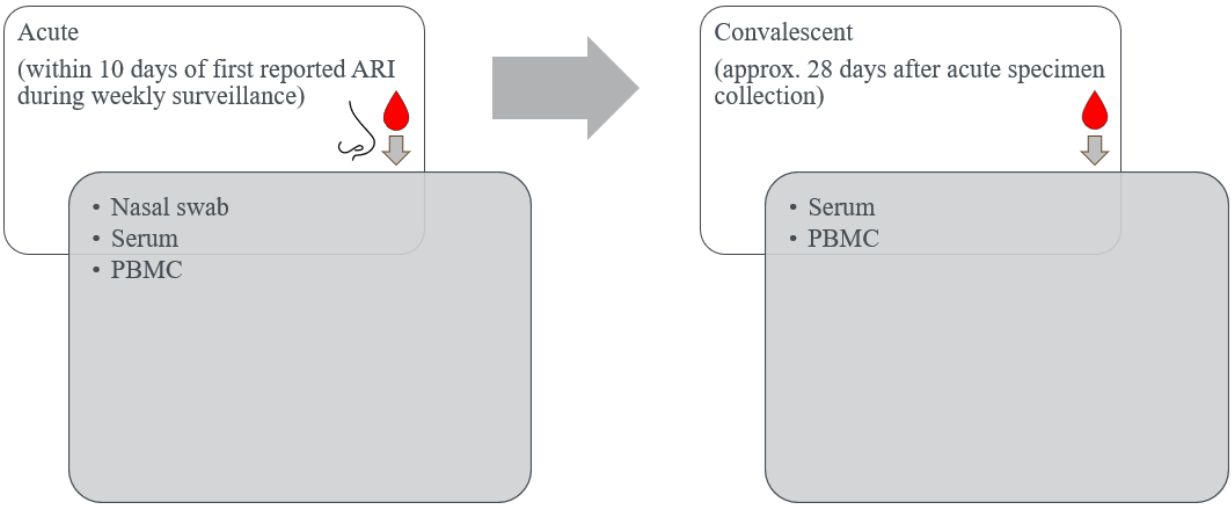
PROTOCOL SUMMARY

Title:	Randomized study of the immunogenicity and duration of antibody response against circulating SARS-CoV-2 variant and influenza viruses following concomitant versus sequential administration of mRNA COVID-19 vaccine and quadrivalent cell culture-based influenza vaccine among children and adults
Phase:	Phase IV
Population:	600 persons aged 18-64 years and 200 children aged 6-11 years for whom a single dose of 2023-24 influenza vaccine and a single dose of mRNA COVID-19 vaccine is indicated as recommended by ACIP
Clinical Sites:	Four study sites: Arizona State University Tempe; University Hospitals of Cleveland; Washington University St. Louis; University of Pittsburgh
Coordinating Site:	Duke University
Study Duration:	12 months total Enrollment: Aug — Oct 2023 Vaccination: Aug — Nov 2023 Blood draws: Aug 2023 — May 2024 Active surveillance for illness: Aug 2023 — May 2024 Laboratory testing: Nov 2023 — Jul 2024
Participant Duration and Number of Participants:	• Up to 10 months (Aug 2023 — May 2024) • 800 total participants (600 aged 18-64 years, 200 aged 6-11 years) at 4 sites (only two sites will enroll children aged 6-11 years)
Randomization:	Adults aged 18-64 years, 2:1:1 permuted block randomization i. Concomitant ccIIV4 + COVID-19 mRNA (day 0) [n = 300] ii. Sequential ccIIV4 (day 0), COVID-19 mRNA (day 28) [n = 150] iii. Sequential COVID-19 mRNA (day 0), ccIIV4 (day 28) [n = 150]

	Children aged 6-11 years, 2:1:1 permuted block randomization i. Concomitant ccIIV4 + Moderna COVID-19 mRNA (day 0) [n = 100] ii. Sequential ccIIV4 (day 0), Moderna COVID-19 mRNA (day 28) [n = 50] iii. Sequential Moderna COVID-19 mRNA (day 0), ccIIV4 (day 28) [n = 50]
	
Description of Study Procedures:	This study is a randomized immunogenicity study in an enrolled cohort with active surveillance for acute respiratory illness (ARI). During this study, participants will be randomly assigned to receive an approved quadrivalent influenza vaccine (Flucelvax [ccIIV4]) and mRNA COVID-19 vaccine (Moderna) either concomitantly (given in contralateral arms) or sequentially (28 days apart). Blood samples from participants will be collected for measurement of biomarkers of immune response at baseline (visit 1; day 0), post-vaccination 1 (visit 2; day 28), post-vaccination 2 (visit 3; day 56), and post-season (visit 4; day 180). Serum and peripheral blood mononuclear cells (PBMC) and plasma (target to collect PBMC and plasma samples at every visit from a subset of approximately 250 participants; 200 adults and 50 children) samples will be isolated from whole blood and tested for biomarkers of vaccine immunogenicity, and duration of antibody responses.

	Participants will receive electronic surveys via email or text message three days after each vaccination asking about vaccine reactogenicity and weekly asking about change in health status and new ARI symptoms; those reporting illness may be asked to provide a respiratory swab for laboratory testing for influenza and SARS-CoV-2 and up to 2 additional blood draws (acute [< 10 days after symptom onset] and convalescent [28 days after acute visit if lab-confirmed positive for influenza or SARS-CoV-2]).
Specimen Collection:	Adults aged 18-64 years, n = 600 i. Sera [n = 600] ii. PBMC [target n = 200] Children aged 6-11 years, n = 200 i. Sera [n = 200] ii. PBMC [target n = 50]
All participants will present for 4 visits: 	

If reporting symptoms, participants may be asked to present for two additional visits:



Objectives:	Primary Objective: i. Determine baseline and post-vaccination influenza antibody titers by hemagglutination inhibition (HAI) (in a subset) against ccIIV4 vaccine antigens following concomitant administration with mRNA vaccine or alone Exploratory Objective: ii. Determine baseline and post-vaccination influenza antibody titers by microneutralization (MN) (in a subset) against ccIIV4 vaccine antigens following concomitant administration with mRNA vaccine or alone iii. Determine baseline and post-vaccination SARS-CoV-2 antibody binding titers and ACE2 binding inhibition and viral neutralization, on a subset of specimens, against Moderna mRNA COVID-19 vaccine antigens following concomitant administration with ccIIV4 or alone iv. Determine duration and rate of waning of influenza HAI and MN antibody titers against ccIIV4 vaccine antigens and circulating influenza viruses v. Determine duration of and rate of waning of SARS-CoV-2 binding antibody and neutralization titers (in a subset of specimens) against Moderna mRNA COVID-19 vaccine antigens and circulating SARS-CoV-2 variants

	vi. Determine influenza HAI and MN antibody titers against circulating influenza viruses during baseline and post-vaccination vii. Determine SARS-CoV-2 antibody binding titers and viral neutralization titers (in a subset of samples) against circulating SARS-CoV-2 variants during baseline and post-vaccination viii. Compare the frequencies of antigen-specific T and B cells utilizing high-dimensional flowcytometric analysis of innate and adaptive immune cell phenotype characterization using fluorochrome-conjugated antigen tags, and intracellular cytokine staining to 2023-24 ccIIV4 and mRNA COVID-19 vaccines administered either concomitantly or sequentially, 28 days apart ix. Compare transcriptome, B cell receptor (BCR) and T cell receptor (TCR) diversity of antigen-specific cells and assess serological antibody repertoire (clonotypes) using 10X genomics platform, and liquid chromatography/mass spectroscopy in a subset of participants x. Investigate the waning of antigen-specific B cells and antibody clonotypes in a subset of participants xi. Describe safety of concomitant versus sequential administration of ccIIV4 and mRNA COVID-19 vaccination
Outcome Measures:	Primary Outcome Measures: i. The percent seroconversion and seroprotective titer, the geometric mean titer (GMT), and geometric mean fold rise (GMFR) in titer for each vaccination group across HAI titers for each 2023-24 influenza vaccine antigen Exploratory Outcome Measures: ii. The percent seroconversion and seroprotective titer, the geometric mean titer (GMT), and geometric mean fold rise (GMFR) in titer for each vaccination group across MN titers for each 2023-24 influenza vaccine antigen iii. GMT for SARS-CoV-2 binding antibody titer and viral neutralization titer (for a subset of samples) pre- and post-vaccination with mRNA COVID-19 vaccine iv. GMT and geometric mean fold change in HAI or MN titers at baseline, post-vaccination, mid-season, and post-season against 2023-24 circulating influenza virus and influenza vaccine antigens

	v. GMT and geometric mean fold change for SARS-CoV-2 binding antibody and the neutralization titer (for a subset of samples) at baseline, post-vaccination, mid-season, and post-season against 2023-24 circulating SARS-CoV-2 virus and mRNA COVID-19 vaccine antigens vi. Percent seroconversion and seroprotective titer, GMT, GMFR in HAI or MN titers at baseline and post-vaccination against 2023-24 circulating influenza virus antigens vii. GMT and GMFR for SARS-CoV-2 binding antibody and the neutralization titer (for a subset of samples) at baseline and post-vaccination against 2023-24 circulating SARS-CoV-2 virus antigens viii. The frequency of antigen-specific T and B cells to influenza and SARS-CoV-2 at baseline, post-vaccination, mid-season and post-season, high-dimensional flowcytometric analyses of innate and adaptive immune cells, and ELISPOT evaluation for SARS-CoV-2 memory B cells ix. BCR and TCR diversity and clonal dominance (antibody clonotypes) of serological repertoire in a subset of participants x. The frequency of antigen-specific B cells and clonal dominance (antibody clonotypes) to influenza and SARS-CoV-2 at baseline, post-vaccination, mid-season and post-season in a subset of participants xi. Number and percent of participants reporting fever or injection site and systemic reactions during the first 3 days following each vaccination and a change in health requiring hospital admission during the study period
Estimated Time to Complete Enrollment:	3 months: Aug — Oct 2023

1 TITLE OF PROJECT

Randomized study of the immunogenicity and duration of antibody response against circulating SARS-CoV-2 variant and influenza viruses following concomitant versus sequential administration of mRNA COVID-19 vaccine and quadrivalent cell culture-based influenza vaccine among children and adults

Short Title: Comparative immunogenicity of concomitant vs sequential mRNA and influenza vaccination

1.1 Study Sponsor

Influenza Division, U.S. Centers for Disease Control and Prevention

1.2 Investigators

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2 ABBREVIATIONS

ACE2	Angiotensin Converting Enzyme 2
ARI	Acute Respiratory Illness
BCR	B cell receptor
ccIIV4	Flucelvax Quadrivalent
GMT	Geometric Mean Titers
GMFR	Geometric Mean Fold Rise
HAI	Hemagglutination Inhibition
IRB	Internal Review Board
LAR	Legally Authorized Representative
MN	Microneutralization
PBMC	Peripheral Blood Mononuclear Cells
RT-PCR	Reverse-Transcription Polymerase Chain Reaction
SAE	Serious Adverse Event
TCR	T cell receptor

3 BACKGROUND

3.1 Protocol Summary

This protocol describes an immunologic study of response to influenza and SARS-CoV-2 vaccination across four of the US Influenza Vaccine Effectiveness (Flu VE) Network study sites. This protocol meets the objectives of component D in the Flu VE Network Cooperative Agreement (IP16-004), including enrollment of eligible persons into a cohort, administration of a licensed or authorized under emergency use authorization (EUA) mRNA COVID-19 vaccine (Moderna COVID-19 Vaccine) and licensed quadrivalent cell culture-based influenza vaccine (Flucelvax; ccIIV4), collection and testing of blood specimens, administration of enrollment surveys, protection of personally identifiable information, transfer of a HIPAA-limited dataset to the Network Coordinating Center (NCC) and CDC, and creation of a coded individual-level dataset according to a data management agreement.

3.2 Protocol Background and Rationale

Current guidance concerning administration of U.S.-approved or -authorized COVID-19 vaccines indicates that these vaccines may be given with influenza vaccines.[1]

In accordance with general best practices, routine administration of all age-appropriate doses of vaccines concomitantly is recommended for children, adolescents, and adults for whom no specific contraindications exist at the time of the healthcare visit. Providers should be aware of the potential for increased reactogenicity with concomitant administration and should consult updated CDC guidance as more information becomes available.

Extensive experience with non-COVID-19 vaccines has demonstrated that immunogenicity and adverse event profiles are generally similar when vaccines are administered concomitantly as when they are administered alone.

Studies that compared coadministration of COVID-19 vaccines and seasonal influenza vaccines with separate administration of these vaccines found similar levels of immunogenicity and similar or slightly higher reactogenicity;[2-4] no specific safety concerns were identified.

During the 2023-2024 influenza season, CDC plans a randomized influenza vaccine immunogenicity study to compare immune responses to concomitant vs sequential administration of licensed or authorized vaccines for COVID-19 and influenza in select age groups. For future seasons, immunogenicity studies may enroll different age groups each season depending on the vaccines to be compared. To study immune responses to vaccination, each study site will recruit and enroll up to 250 participants who are eligible to receive specific influenza and COVID-19 vaccines and for whom the vaccines are recommended. Study sites will implement common protocols developed in collaboration with CDC. Data will be combined across sites so that all sites will contribute to a total anticipated sample size of 800 participants for comparisons of immune responses by vaccine administration schedule. Investigators will collect, process, store, and ship sera, plasma, and PBMC specimens to a designated CDC laboratory for testing. Some additional specimen testing may be performed by a CDC-approved site.

Investigators will participate in cross-site decisions on specific influenza and COVID-19 vaccines to be administered, populations to be examined, and interpretation of influenza vaccine response findings. Enrolled participants may either reside or work in settings that facilitate active weekly surveillance for ARI and influenza virus and SARS-CoV-2 infection. To improve representation of specific population groups, study sites may use enrollment quotas for participant demographics including race and ethnicity. To identify influenza virus and SARS-CoV-2 infections, participants will respond to weekly automated electronic surveys to report new ARI symptoms and test results, if applicable. Participants may have the opportunity to provide specimens for standard point of care respiratory testing and up to 2 additional blood draws (at acute illness and 28 days after PCR-confirmed influenza or SARS-CoV-2 infection) if they report qualifying ARI symptoms during weekly surveillance. Participants will provide scheduled blood

samples at enrollment (before vaccination) and at several timepoints after vaccination to measure and monitor humoral and cell-mediated immune (CMI) responses to vaccination. Participants will respond to an automated electronic survey 3 days following each vaccination visit to report the occurrence of fever, injection site reactions, systemic reactions, the severity of reactions and their impact on health. The weekly automatic electronic survey will also request participants to report the occurrence of any change in health requiring hospitalization.

3.3 Vaccines Background and Rationale

3.3.1 COVID-19 Vaccines

COVID-19 vaccines available in the United States are effective at protecting people from getting seriously ill, being hospitalized, and dying from SARS-CoV-2. U.S.-licensed or authorized under EUA mRNA COVID-19 vaccine used in this study will contain the recommended composition to be used as a booster dose at the time of enrollment. Currently, CDC recommends one bivalent vaccine dose for everyone aged 6 years and older that have not previously been vaccinated or have only received monovalent doses if it has been at least 2 months since their last dose.[5]

Two COVID-19 vaccine manufacturers, Pfizer and Moderna, have developed updated (bivalent) COVID-19 boosters. The updated (bivalent) boosters are called “bivalent” because they protect against both the original or ancestral virus that causes COVID-19 and the Omicron variant BA.4 and BA.5. Primary series doses and previous boosters are called “monovalent” because they were designed to protect against the original virus that causes COVID-19. They also provide some protection against Omicron, but not as much as the updated (bivalent) boosters. The virus that causes COVID-19 has changed over time. The different versions of the virus that have developed over time are called variants. As variants continue to evolve it remains unclear at this time what the composition of future booster doses of COVID-19 vaccine. The latest Moderna COVID-19 vaccine will be administered in this trial.

3.3.2 Influenza Vaccines

U.S.-licensed 2023-2024 cell culture-based influenza vaccine used in this study will contain components from four influenza viruses antigenically similar to those recommended by FDA for cell culture–based vaccines: an influenza A(H1N1)pdm09 virus; an influenza A(H3N2) virus; an influenza B/Victoria lineage virus; and an influenza B/Yamagata lineage virus. Most commonly, seasonal influenza vaccines are produced using egg-grown viral isolates; however, this approach could lead to individual’s receiving a boost in antibody protection against egg-adapted antigens which may vary from circulating influenza viruses.[1-4] Previous work in adults has suggested that vaccination with cell culture-based derived vaccines could improve immune response to circulating influenza viruses compared to traditional egg-based vaccines.[6-8]

A cell culture-based influenza vaccine, Flucelvax Quadrivalent (ccIIV4), will be administered in this trial.[9] ccIIV4 may be administered concomitantly or sequentially with other inactivated vaccines. Injectable vaccines that are given concomitantly should be administered at separate anatomic sites.

Standard-dose, nonadjuvanted cell culture-based vaccine, Flucelvax Quadrivalent (ccIIV4) contains 15 µg of HA per vaccine virus in a 0.5-mL dose. ccIIV4 is approved for persons aged ≥6 months. Egg-based and cell culture-based vaccines differ in the substrate in which reference vaccine viruses supplied to the manufacturer are propagated in quantities sufficient to produce the needed number of doses of vaccine. For Flucelvax Quadrivalent, reference vaccine viruses are propagated in Madin-Darby canine kidney cells instead of eggs.

4 STUDY OBJECTIVES AND OUTCOME MEASURES

OBJECTIVE	OUTCOME
4.1 Primary	
Determine baseline and post-vaccination influenza antibody titers by hemagglutination inhibition (HAI), in a subset of samples, against vaccine antigens following concomitant administration with mRNA vaccine or alone	i. Proportion of participants in each vaccination group with a seroprotective HAI (in a subset of samples) titer (≥1:40) pre- and post-ccIIV4 immunization for each 2023-24 influenza vaccine antigen ii. Proportion of participants in each vaccination group achieving HAI (in a subset of samples) seroconversion (a titer ≥1:40 following ccIIV4 if the baseline titer is <1:10 or a four-fold rise in titer if the baseline titer is >1:10) following ccIIV4 vaccination for each 2023-24 influenza vaccine antigen iii. HAI (in a subset of samples) GMT for each 2023-24 influenza vaccine antigen pre- and post-ccIIV4 in each vaccination group iv. GMFR in HAI (in a subset of samples) titers for each 2023-24 influenza

	vaccine antigen in each vaccination group
4.2 Exploratory	
Determine baseline and post-vaccination influenza antibody titers by microneutralization (MN), in a subset of samples, against vaccine antigens following concomitant administration with mRNA vaccine or alone	i. Proportion of participants in each vaccination group with a seroprotective MN (in a subset of samples) titer (≥1:40) pre- and post-ccIIV4 immunization for each 2023-24 influenza vaccine antigen ii. Proportion of participants in each vaccination group achieving MN (in a subset of samples) seroconversion (a titer ≥1:40 following ccIIV4 if the baseline titer is <1:10 or a four-fold rise in titer if the baseline titer is >1:10) following ccIIV4 vaccination for each 2023-24 influenza vaccine antigen iii. HAI (in a subset of samples) MN for each 2023-24 influenza vaccine antigen pre- and post-ccIIV4 in each vaccination group i. GMFR in MN (in a subset of samples) titers for each 2023-24 influenza vaccine antigen in each vaccination group
Determine baseline and post-vaccination SARS-CoV-2 antibody binding titers and ACE2 binding inhibition and viral neutralization, on a subset of specimens, against mRNA COVID-19 vaccine antigens following concomitant administration with ccIIV4 or alone	i. GMT for SARS-CoV-2 binding antibody titer and viral neutralization titer (for a subset of samples) pre- and post-vaccination with mRNA COVID-19 vaccine
Determine duration and rate of waning of influenza HAI and MN antibody titers	i. HAI and MN GMT for each 2023-24 influenza vaccine and circulating influenza virus antigens at visit 1, visit

against ccIIV4 vaccine antigens and circulating influenza viruses	2, visit 3, and visit 4 for each vaccination group ii. Geometric mean fold change in HAI and MN titers for each vaccination group from visit 1 to visit 2, visit 3, and visit 4 for each 2023-24 influenza vaccine and circulating influenza virus antigen
Determine duration of and rate of waning of SARS-CoV-2 antibody titers and viral neutralization titers, on a subset of samples, against mRNA COVID-19 vaccine antigens and circulating SARS-CoV-2 variants	i. SARS-CoV-2 binding antibody and viral neutralization GMT for each mRNA COVID-19 vaccine and circulating SARS-CoV-2 virus antigens at visit 1, visit 2, visit 3, and visit 4 for each vaccination group ii. Geometric mean fold change in SARS-CoV-2 and viral neutralization (in a subset of samples) titers for each vaccination group from visit 1 to visit 2, visit 3, and visit 4 for each COVID-19 Moderna mRNA COVID-19 vaccine and circulating SARS-CoV-2 virus antigen
Determine influenza HAI and MN antibody titers against circulating influenza viruses during baseline and post-vaccination or post-infection	i. Proportion of participants in each vaccination group with a seroprotective titer (≥1:40) pre- and post-ccIIV4 immunization (visits 1-3) and/or pre- and post-influenza infection (acute and convalescent visits), if applicable, for each 2023-24 circulating influenza virus antigen ii. Proportion of participants in each vaccination group achieving seroconversion (a titer ≥1:40 following ccIIV4 if the baseline titer is <1:10 or a four-fold rise in titer if the baseline titer is >1:10) following ccIIV4 vaccination (visit 1 to visits 2-3) and/or influenza

	infection (acute to convalescent visits), if applicable, for each 2023-24 circulating influenza virus antigen iii. HAI and MN GMT for each 2023-24 circulating influenza virus antigen pre- and post-ccIIV4 (visits 1-3) and/or pre- and post-influenza infection (acute and convalescent visits), if applicable, in each vaccination group iv. GMFR in HAI or MN titers for each vaccination group following ccIIV4 vaccination (visit 1 to visits 2-3) and/or influenza infection (acute to convalescent visits), if applicable, for each 2023-24 circulating influenza virus antigen
Determine SARS-CoV-2 antibody binding titers and viral neutralization titers (in a subset of samples) against circulating SARS-CoV-2 variants during baseline and post-vaccination	i. SARS-CoV-2 binding antibody and viral neutralization (for a subset of samples) titers GMT at pre- and post-vaccination (visits 1-3) and/or pre- and post-SARS-CoV-2 infection (acute and convalescent visits), if applicable, against 2023-24 circulating SARS-CoV-2 virus antigens ii. GMFR in SARS-CoV-2 binding antibody and viral neutralization (for a subset of samples) titers following mRNA COVID-19 vaccination (visit 1 to visits 2-3) and/or SARS-CoV-2 infection (acute to convalescent visits), if applicable, against 2023-24 circulating SARS-CoV-2 virus antigens
Compare the frequencies of antigen-specific T and B cells utilizing conventional ELISPOT assays, high-dimensional flowcytometric analysis of innate and adaptive immune cell phenotype	i. The frequency of antigen-specific T and B cells to influenza and SARS-CoV-2 at visit 1, visit 2, visit 3, visit 4, and acute and convalescent visits, if applicable

characterization using fluorochrome-conjugated antigen tags, and intracellular cytokine staining to 2023-24 ccIIV4 and mRNA COVID-19 vaccines administered either concomitantly or sequentially, 28 days apart	ii. High-dimensional flowcytometric analyses of innate and adaptive immune cells iii. ELISPOT evaluation for SARS-CoV-2 memory B cells
Compare transcriptome, B cell receptor (BCR) and T cell receptor (TCR) diversity of antigen-specific cells and assess serological antibody repertoire (clonotypes) using 10X genomics platform, and liquid chromatography/mass spectroscopy in a subset of participants	i. BCR and TCR diversity and clonal dominance (antibody clonotypes) of serological repertoire in a subset of participants
Investigate the waning of antigen-specific B cells and antibody clonotypes in a subset of participants	i. The frequency of antigen-specific B cells and clonal dominance (antibody clonotypes) to influenza and SARS-CoV-2 at visit 1, visit 2, visit 3, visit 4, and acute and convalescent visits (if applicable) in a subset of participants
Describe safety of concomitant versus sequential administration of ccIIV4 and mRNA COVID-19 vaccination	ii. Number and percent of participants reporting fever or injection site and/or systemic reactions during the first 3 days following each vaccination visit stratified by severity and, for those reporting symptoms, their impact on health iii. Number and percent of participants reporting a change in health status requiring hospital admission (including a description of the event) during the study period

5 METHODS

5.1 Main study design

This study is a randomized comparative immunogenicity study in an enrolled cohort. During this study, participants will be randomly assigned to receive an approved quadrivalent cell culture-based influenza vaccine (ccIIV4) and mRNA COVID-19 vaccine (Moderna) either concomitantly or sequentially, 28 days apart. Participants (aged 6-11 years and 18-64 years) will be enrolled in the 2023-2024 influenza season. Only two of the sites (Washington University in St. Louis and University Hospitals Cleveland Medical Center) will enroll children aged 6-11 years. Individuals will be enrolled who have not received their 2023-2024 influenza vaccine and have not received any COVID-19 vaccine in the preceding 6 months (i.e. at least 6 months have passed since their last COVID-19 vaccination).

Participants will be enrolled if one dose of 2023-2024 influenza vaccine and a single booster dose of mRNA COVID-19 vaccine are indicated for current vaccination according to ACIP recommendations. To qualify for the study, children aged 8 years and younger must only need to receive a single dose of ccIIV4 per ACIP recommendations for the 2023-2024 season. mRNA COVID-19 vaccines recommended by CDC and/or authorized or approved by the FDA at the time of enrollment may include updated SARS-CoV-2 viral components which may be included in this study if authorized or approved by the FDA and recommended by the CDC.

Demographic and health data (including influenza and COVID-19 vaccination and infection history) will be collected upon enrollment and verified after enrollment via electronic medical record extraction. Enrolled participants will be randomized to one of the following interventions (2:1:1) using a permuted block method stratified by age group (aged 6-11 years and 18-64 years) and site: (i) concomitant administration of the mRNA COVID-19 vaccine (Moderna) and quadrivalent influenza vaccine (ccIIV4); (ii) sequential administration of the quadrivalent influenza vaccine (ccIIV4) at Visit 1 (day 0) and the mRNA COVID-19 vaccine (Moderna) at Visit 2 (day 28); (iii) sequential administration of the mRNA COVID-19 vaccine (Moderna) at Visit 1 (day 0) followed by the quadrivalent influenza vaccine (ccIIV4) at Visit 2 (day 28). Whole blood samples to isolate sera, PBMC and plasma (target to collect PBMC and plasma samples at every visit from a subset of 250 participants; 200 adults and 50 children) will be collected prior to vaccination administration at Visit 1 (day 0) and Visit 2 (day 28). Participants will not be blinded to vaccine group and will receive two vaccines in separate limbs (for the concomitant vaccination [group i]) or one vaccine at Visit 1 (day 0) and a second vaccine at Visit 2 (day 28) (for sequential vaccination [groups ii and iii]) in an unblinded manner.

Participants will provide a blood sample for isolation of serum, PBMC, and plasma (target to collect PBMC and plasma samples at every visit from a subset of 250 participants; 200 adults and 50 children) at Visit 3 (day 56; post-vaccination 2) and Visit 4 (day 180; end of local flu circulation). If participants exhibit ARI during the study period, they may be asked to present for

an acute (<10 days after symptom onset) and convalescent (28 days after acute visit) visit to provide a nasal swab for viral testing (acute visit only) and blood sample for isolation of serum, PBMC, and plasma (acute and convalescent visit for those with positive viral molecular test for influenza or SARS-CoV-2). Influenza virus and SARS-CoV-2 serologic and cellular immunity testing will be conducted at CDC and/or a CDC-approved site.

5.2 Laboratory Studies

Blood samples from participants will be collected for measurement of biomarkers of immune response at Visit 1 (day 0; baseline), Visit 2 (day 28; post-vaccination 1), Visit 3 (day 56; post-vaccination 2), and Visit 4 (day 180; post-season). Sera, PBMC, and plasma samples will be tested for biomarkers of vaccine immunogenicity, duration of antibody responses, and immune response to circulating SARS-CoV-2 variants and influenza virus strains.

5.2.1 Influenza Antibody Assays

5.2.1.1 Influenza Hemagglutination Inhibition and Microneutralization Assays

Sera collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), acute visit (<10 days after symptom onset) and convalescent visit (28 days after Acute Visit) will be stored at -20 C or colder (collection and storage described in detail in Lab Appendix B) and tested for influenza immunologic assays including hemagglutination inhibition (HAI) and microneutralization (MN) antibody titers. HAI and MN antibody titers will be compared between groups receiving COVID-19 and ccIIV4 concomitantly or alone for each of the four 2023-2024 recommended influenza vaccine strains for the cell culture-based vaccine (ccIIV4), as well as against cell culture-based propagated circulating influenza viruses identified during the 2023-2024 influenza season. Participants will not receive individual HAI or MN antibody titer results; these are not routinely used in clinical practice.

5.2.1.2 Influenza Cell Mediated Immunity Assays

PBMCs and plasma will be isolated from whole blood samples collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), Acute Visit (<10 days after symptom onset) and Convalescent Visit (28 days after Acute Visit) to be stored for influenza immunologic assays. PBMCs will be stored in liquid nitrogen and plasma will be stored at -80 C (collection and storage described in detail in Lab Appendix C). PBMC and plasma isolates will be shipped to CDC and the CDC influenza immunology laboratory will transfer the samples to the identified testing laboratories. PBMCs will be tested to assess the frequencies of H1-, H3-, B- (Yamagata and Victoria) specific memory B cells as well as Groups 1, 2 and B stem-specific memory B cells using fluorochrome-conjugated influenza HA and Stem tags. In addition, the frequencies of follicular helper T cells and antigen-specific T cells post-stimulation with influenza

viral antigens or influenza viruses will be assessed by high-dimensional flowcytometry and intracellular cytokine staining. PBMCs and plasma from a subset of participants will be tested for transcriptome, BCR, TCR and antibody repertoire using single cell genomics with 10X Genomic platform and plasma antibody repertoire with LC/MS.

5.2.2 SARS-CoV-2 Antibody Assays

5.2.2.1 Quantitative SARS-CoV-2 antibody binding assessment

Sera isolated from whole blood samples collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), acute Visit (<10 days after symptom onset) and convalescent Visit (28 days after Acute Visit) will be stored and tested for SARS-CoV-2 binding antibody using the Meso Scale Discovery (MSD) assay. Sera will be assayed for the presence of SARS-CoV-2 antibody at CDC. Participants will not receive individual COVID-19 antibody assay results; these are not routinely used in clinical practice.

5.2.2.2 Quantitative SARS-CoV-2 assessments

Sera will be isolated from whole blood samples collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), Acute Visit (<10 days after symptom onset) and Convalescent Visit (28 days after Acute Visit) to be stored for sera COVID-19 neutralization assays. ACE2 binding inhibition in sera will be measured using MSD and percent inhibition compared between groups receiving COVID-19 and ccIIV4 simultaneously or sequentially for each variant tested. COVID-19 neutralizing antibody titers will be compared in a subset of samples between groups receiving COVID-19 and ccIIV4 simultaneously or sequentially for each variant tested. Participants will not receive individual COVID-19 antibody titer results; these are not routinely used in clinical practice.

5.2.2.3 SARS-CoV-2 Cell Mediated Immunity Assays

PBMCs and plasma isolated from whole blood samples collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), Acute Visit (<10 days after symptom onset) and Convalescent Visit (28 days after Acute Visit) will be stored and tested for SARS-CoV-2 immunologic assays to evaluate the memory response to SARS-CoV-2. PBMCs will be evaluated to detect the presence of SARS-CoV-2 specific CD4+ and CD8+ memory T cells by high-dimensional flowcytometry and intracellular cytokine staining. The memory B cell population specific to SARS-CoV-2 antigens will be evaluated by ELISPOT. Participants will not receive individual COVID-19 antibody results; these are not routinely used in clinical practice.

5.2.3 Future studies

In addition to the specified analyses described thus far, there may be other tests or assays, including antibody binding assays, that have yet to be identified that may be important for interpreting our study findings or of relevance to vaccine outcomes. Additional laboratory assays may test for antibodies against other bacteria or viruses, markers of inflammation, or used in research on the health of the participants. Specimens banked for use in other studies will be linked to information (including identifying information) that participants provided to the study. Participants/participant's legally authorized representative (LAR) must agree to potential future use of samples in order to be in the study. No human genetic tests will be performed on samples collected in this study. Because it is unknown if future testing will be of any utility, results of future testing will not be provided.

5.3 Study Enrollment and Withdrawal

5.3.1 Participant Inclusion Criteria

Participants who meet all of the following criteria and consent to randomization to one of three vaccine scheme groups will be eligible to participate in this interventional study:

i. Healthy children aged 6-11 years and healthy adults aged 18-64 years that have not received the current season's influenza vaccine or a mRNA COVID-19 vaccine in the past 6 months and have previously received at least a single dose of any US approved or authorized COVID-19 vaccine
ii. English or Spanish literate
iii. Email or text message capability for weekly follow-up
iv. Intention of receiving influenza vaccine and mRNA COVID-19 vaccine based on ACIP-CDC guidelines
v. Willing to provide written/electronic informed consent
vi. Intention of being available for entire study period and able to complete all relevant study procedures, including follow-up phone calls and clinic visits

5.3.2 Participant Exclusion Criteria

Participants who meet any of the following criteria or do not consent to receiving both vaccines at enrollment, or one 28 days later, will not be eligible to participate in this study:

i. Self-reported COVID-19 infection within 3 months prior to enrollment
ii. Received COVID-19 vaccine within 6 months prior to enrollment

iii. Received influenza vaccine during the respective influenza season in which they are being enrolled

iv. < 9 years of age and recommended to receive two doses of IIV4 during the respective influenza season in which they are being enrolled

v. History of severe allergic reaction after a previous dose of any influenza or COVID-19 mRNA vaccine; or to an influenza or COVID-19 mRNA vaccine component

vi. Receipt of any licensed vaccine within 6 weeks prior to enrollment in this study or planning receipt of any vaccines within 4 weeks after the receipt of the second vaccine dose administered during study procedures

vii. Has an immunocompromising condition or taking immunosuppressive medication*

* Received oral, intramuscular or intravenous systemic immunosuppressants, or immune modifying drugs for >14 days in total within 6 months prior to any study vaccine dose (for corticosteroids ⩾ 20 mg/day of prednisone equivalent).

** Note: Topical medications are allowed

viii. Received immunoglobulin, SARS-CoV-2 immunoglobulin, SARS-CoV-2 monoclonal antibody, or blood-derived products, within 3 months prior any study vaccine dose.

ix. History of Guillain-Barré syndrome

x. History of myocarditis or pericarditis

xi. History of multisystem inflammatory syndrome in children (MIS-C) or adults (MIS-A)

xii. Currently pregnant, planning to become pregnant within the first three months of the study per participant self-report or likely to be pregnant per screening criteria

xiii. Bleeding disorder diagnosed by a healthcare provider or bleeding difficulties with intramuscular injections or blood draws.

xiv. Has injury or other reason why deltoid site on both arms cannot be used for vaccinations

xv. Any condition which, in the opinion of the investigators, may pose a health risk to the participant or interfere with the evaluation of the study objectives

5.3.3 Temporary Delay Criteria (Visits 1 and 2)

Participants who meet any of the following criteria will be subject to temporary delay in protocol:

i. History of febrile illness (> 100.0°F or 37.8°C) within the past 72 hours prior to vaccine administration

5.3.4 Recruitment

Physical distancing and other COVID-19 control measures will be incorporated into all study activities according to state and local guidance. The study will use multiple recruitment methods to reach potential participants within the study catchment area. These recruitment methods will be informed by site experience with their local populations. Sites will be asked to make efforts to engage, partner with, and recruit from communities of color to recruit a participant population whose self-reported race and ethnicity distribution approximates the racial and ethnic composition of the United States based on US Census data. Each site will aim to enroll 250 participants.

5.3.4.1 University of Pittsburgh

Directly approaching potential participants (in-person)

Medical records will be reviewed by staff who have legitimate access to this information. For example, pediatric GAP nurses already have privileges to see and access data at Pediatric care sites. The study script would be read to a potentially eligible participant who could decide if they want to hear more about the study.

Email/Listserv/Electronic Mailing List, Letters sent to potential participants, telephone scripts, Registries (remote initial contact)

Recruitment will include participants from prior research studies (STUDY19040242, STUDY22040177, STUDY22100188) or registries (STUDY20040088 and Pitt+Me) who agreed to be contacted regarding research participation as well as mailing lists through the University of Pittsburgh employees (Read Green employee newsletter email, for example).

If contact is by mail (prior research studies), a study recruitment letter will be sent to potential participants meeting the inclusion criteria. In the recruitment letter, we will briefly describe study activities and provide contact information for individuals interested in potential participation.

If contact is made by text, email, phone, or voicemail (these three methods will be used for those both previously enrolled and from the registry who have not previously enrolled), a brief message will be used and a script is attached.

Those who agree to be screened after the study is described to them can be transferred to the research assistant for screening and potential enrollment. All potential participants referred to our study will undergo standard screening by the study team using the REDCap screening survey and will not require medical record review to determine eligibility.

Information collected during the screening process on participants who are deemed ineligible, regardless of the recruitment method, will be discarded.

5.3.4.2 Washington University in St. Louis

Washington University in St. Louis will recruit participants from their study registry, Volunteers for Health, via emails and social media posts.

5.3.4.3 University Hospitals Cleveland Medical Center

University Hospitals Cleveland will recruit from participants that were enrolled in previous vaccine trials, employees, and via Facebook advertising and brochures.

5.3.4.4 Arizona State University, Tempe

Arizona State University will recruit local student and faculty population through study flyers. Valleywise Health will recruit employees via study flyers and the intranet. Phoenix Children's Hospital will approach and recruit patients presenting for seasonal influenza vaccination or mRNA COVID-19 vaccination and via study flyers.

5.3.5 Enrollment

Enrollment in this study will begin once 2023-24 seasonal influenza vaccinations become available, anticipated August 2023. Enrolled participants will be randomized to one of the following three interventions (2:1:1) using a permuted block method stratified by site: (i) concomitant administration of the mRNA COVID-19 vaccine and quadrivalent influenza vaccine (ccIIV4); (ii) sequential administration of the quadrivalent influenza vaccine (ccIIV4) at Visit 1 (day 0) and the mRNA COVID-19 vaccine at Visit 2 (day 28); (iii) sequential administration of the mRNA COVID-19 at Visit 1 (day 0) followed by the quadrivalent influenza vaccine (ccIIV4) at Visit 2 (day 28).

Individuals will be recruited to participate in the study using site-specific methods which may include virtual and/or in-person recruitment. Potentially eligible individuals will be directed to complete the eligibility screening and consent processes to determine whether inclusion/exclusion criteria are met. All individuals who are eligible for the study will view a study description (purposes and procedures) electronically and will be asked to read, sign, and date the written/electronic informed consent and assent, if applicable, forms . The written/electronic informed consent form will explain study details, risks, and benefits and emphasize the voluntary nature of participation. Participants will be directed to contact study staff with questions.

Individuals will be enrolled in the study if they meet inclusion criteria and provide written/electronic informed consent and assent, if applicable. After completing the written/electronic informed consent/assent process, participants will complete an enrollment survey that collects information about demographic characteristics, medical history, and influenza and COVID-19 vaccination history. Enrolled participants will then provide whole blood to isolate sera, PBMC and plasma samples (target to collect PBMC and plasma samples at

every visit from a subset of 250 participants; 200 adults and 50 children) and receive a mRNA COVID-19 vaccine and/or quadrivalent influenza vaccine (ccIIV4) based on their intervention randomization. Study staff will schedule remaining study visits for sample collection (Visits 2, 3, and 4) and second vaccination (Visit 2; groups ii and iii).

5.3.6 Participant Withdrawal

Participants may withdraw from the study at any time for any reason. If a participant withdraws from the study after randomization but prior to receipt of the first vaccine dose administered during study procedure, sites will replace that participant in their randomization group to keep a 2:1:1 randomization scheme. Participants who wish to withdraw from the study will contact the study site principal investigator or coordinator to inform them of their decision. Upon withdrawal from the study, the participant's specimens will be destroyed and discarded if requested by study participant. The participant's data (including responses to the enrollment questionnaire, follow-up survey, and any extracted data) will be removed from all study site databases if requested by study participant.

5.4 Study Schedule, Procedures, & Evaluations

5.4.1 Schedule of events and data collection

Persons meeting the proposed eligibility criteria (**Section 5.3.1**) will be recruited. Written/electronic informed consent will be obtained from study participants/participant's legally authorized representative(s) (LAR) prior to conducting any study procedures.

Table 1: Schedule of Events						
Procedure	**Visit 1**	**Follow-up Survey (Reactogenicity)**	**Visit 2**	**Follow-up Survey (Weekly ARI)**	**Visit 3**	**Visit 4**
	Clinic	Phone/Email	Clinic	Phone/Email	Clinic	Clinic
Estimated Study Day (Relative to Visit 1)	**0**	**3 and 31**	**28**	**Weekly**	**56**	**180**
Time Following Visit 1 (Days) [Window]	0	3 [± 3]	28 [28-42]	Weekly		
Time Following Visit 2 (Days)		3 [± 3] (groups ii and iii only)			28 [28-92]	152 [± 30]
Informed consent & Medical Release of Information	X					
Review Eligibility Criteria	X					
Review Temporary Delay Criteria	X		X			
Demographic and Health History	X					
Influenza and COVID-19 Vaccination History	X					
Randomization	X					
Administer ccIIV4 vaccine	Groups i and ii		Group iii			
Administer Moderna COVID-19 vaccine	Groups i and iii		Group ii			
Whole blood collection for sera sample	Up to 10 mL		Up to 10 mL		Up to 10 mL	Up to 10 mL
Whole blood collection for PBMC and plasma sample	Up to 24 mL [adults] or 8 mL [children]		Up to 24 mL [adults] or 8 mL [children]		Up to 24 mL [adults] or 8 mL [children]	Up to 24 mL [adults] or 8 mL [children]
Obtain solicited adverse events at 3 days following Visits 1 and 2		X				

Table 1: Schedule of Events						
Procedure	**Visit 1**	**Follow-up Survey (Reactogenicity)**	**Visit 2**	**Follow-up Survey (Weekly ARI)**	**Visit 3**	**Visit 4**
	Clinic	Phone/Email	Clinic	Phone/Email	Clinic	Clinic
Estimated Study Day (Relative to Visit 1)	**0**	**3 and 31**	**28**	**Weekly**	**56**	**180**
Time Following Visit 1 (Days) [Window]	0	3 [± 3]	28 [28-42]	Weekly		
Time Following Visit 2 (Days)		3 [± 3] (groups ii and iii only)			28 [28-92]	152 [± 30]
Obtain change in health status requiring hospitalization				X		
Obtain ARI information				X		

Visit 1; study day 0 – Screening, Enrollment, Specimen Collection and Vaccination – Clinic Visit

- Review and confirm study eligibility
- Obtain written/electronic informed consent and assent (if applicable, children age 6-11 years) and release of medical record information
- Obtain information on preferred method of contact for follow-up (text or email)
- Obtain demographic and medical history including history of influenza and COVID-19 vaccination via enrollment questionnaire
- Take temperature and review temporary delay criteria
- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**
- Randomize study participant to intervention (2:1:1) using a permuted block method stratified by site:
    i. concomitant administration of the mRNA COVID-19 vaccine and quadrivalent influenza vaccine
    ii. sequential administration of the quadrivalent influenza vaccine at Visit 1 (day 0) and the mRNA COVID-19 vaccine at Visit 2 (day 28)

iii. sequential administration of the mRNA COVID-19 at Visit 1 (day 0) followed by the quadrivalent influenza vaccine at Visit 2 (day 28).

- Administer mRNA COVID-19 vaccine and/or quadrivalent influenza vaccine (Flucelvax [ccIIV4]) based on randomization scheme
- Confirm date of next appointment at Visit 2 (day 28) of study

Follow-up Survey (Reactogenicity); 3 days after vaccine administration (Visits 1 and 2) – Vaccination Reactogenicity and Safety Data – Online Questionnaire

- Automated reminders sent to all participants to complete electronic survey via participant's preferred contact method approximately 3 days after receipt of either vaccine dose administered according to study procedure (day 0 [groups i-iii] and day 28 [groups ii and iii])
- Participants/participant's LAR(s) complete brief online survey regarding vaccine reactogenicity and safety data in the three days after receipt of either vaccine dose administered according to study procedure

Follow-up Survey (ARI); weekly following Visit 1 – Weekly ARI Surveillance and Change in Health Status – Online Questionnaire

- Automated reminders sent to all participants to complete electronic survey via participant's preferred contact method once per week beginning approximately one week after Visit 1 until completion of Visit 4
- Participants/participant's LAR(s) complete brief online survey regarding new ARI episodes and change in health status requiring hospitalization throughout the entire study period
- Participants that report new ARI may provide respiratory specimens for standard point of care respiratory testing and up to 2 additional blood draws (see Table 2: Schedule of Qualifying ARI Events)

Visit 2; study day 28 (28-42) – Specimen Collection and Vaccination 2 (groups ii and iii) – Clinic Visit

Group i: Concomitant administration of mRNA COVID-19 vaccine and quadrivalent influenza vaccine

- Take temperature and review temporary delay criteria
- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**
- Confirm date of next appointment on day 56 of study

Group ii: Sequential administration of quadrivalent influenza vaccine then mRNA COVID-19 vaccine

- Take temperature and review temporary delay criteria
- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**
- Administer mRNA COVID-19 vaccine
- Confirm date of next appointment on day 56 of study

Group iii: Sequential administration of mRNA COVID-19 vaccine then quadrivalent influenza vaccine

- Take temperature and review temporary delay criteria
- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**
- Administer quadrivalent influenza vaccine (Flucelvax [ccIIV4])
- Confirm date of next appointment on day 56 of study

Visit 3; study day 56 (56-120) – Specimen Collection – Clinic Visit

- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**
- Confirm date of next appointment on day 180 of study

Visit 4; study day 180 – Specimen Collection – Clinic Visit

- Collect whole blood for sera sample **(Section 5.5.4.1)** and PBMC and plasma samples (in subset of participants, target n=250; 200 adults and 50 children) **(Section 5.5.4.2)**

Table 2: Schedule of Qualifying ARI Events

Procedure	Acute Visit	Convalescent Visit
	Clinic	Clinic
Estimated Study Day	**< 10 days after symptom onset**	**28 days after Acute Visit**
Time Following Acute Visit (Days) [Window]	0	28 [28-42]
Nasal swab collection for viral testing	X	
Whole blood collection for sera sample	X	X

Table 2: Schedule of Qualifying ARI Events		
Procedure	**Acute Visit**	**Convalescent Visit**
	Clinic	Clinic
Estimated Study Day	**< 10 days after symptom onset**	**28 days after Acute Visit**
Time Following Acute Visit (Days) [Window]	0	28 [28-42]
Whole blood collection for PBMC and plasma sample	X	X

Acute Visit; <10 Days after Symptom Onset – Specimen Collection – Clinic Visit

If a participant reports ARI symptoms at any point during the weekly electronic follow-up survey (ARI), they may be asked to complete an acute visit. These specimens may be drawn at the same time as scheduled visits 1-4.

- Obtain nasal/throat swab **(Section 5.5.4.3)**
- Obtain sera sample **(Section 5.5.4.1)**, PBMC, and plasma sample **(Section 5.5.4.2)**
- Schedule convalescent visit 28 (28-42) days after acute visit for influenza or SARS-CoV-2 positive participants
- Confirm contact information and preferred method of contact for follow-up (telephone, text, or email)

Convalescent Visit, 28 (28-42) days after acute visit – Specimen Collection – Clinic Visit

If a participant tests positive for influenza and/or SARS-CoV-2 at any acute clinic visit, a convalescent visit is required 28 (28-42) days after their acute clinic visit. These specimens may be drawn at the same time as scheduled visits 1-4.

- Obtain sera sample **(Section 5.5.4.1)**, PBMC, and plasma sample **(Section 5.5.4.2)**

5.4.2 Randomization

Permuted block randomization stratified by age group (children aged 6-11 years and adults aged 18-64 years) and site will be used at each site to ensure even allocation to each study arm. At each site, participants will be randomized 2:1:1 to receive: (i) concomitant administration of a mRNA COVID-19 vaccine and a quadrivalent influenza vaccine (ccIIV4), (ii) sequential administration of a quadrivalent influenza vaccine (ccIIV4) at Visit 1 (day 0) and a mRNA COVID-19 vaccine at Visit 2 (day 28), and (iii) sequential administration of a mRNA COVID-19 vaccine at Visit 1 (day 0) and a quadrivalent influenza vaccine (ccIIV4) at Visit 2 (day 28). The project statistician will generate the randomization schemes which will be uploaded to REDCap. Participants will be randomized through REDCap with treatment allocation recorded

on the case report form. In the event that REDCap is unavailable, participants will be manually randomized through the use of sealed envelopes with randomization allocations enclosed. The project statistician will prepare envelopes that will use the same randomization strategy as the primary scheme embedded in REDCap. When an unblinded team member is informed of the age group, they will pull the next envelope in order. To capture the allocation per subject, a separate form in REDCap will be used by the unblinded study personnel to add the assignment. Sites will need to keep a log capturing these instances. The next available sequential study-number will be assigned to each enrolled participant upon study-entry.

5.4.3 Deviation from Protocol

Influenza and COVID-19 vaccine may be administered outside of study window in case of public health emergency or personal exposure.

5.5 Data Collection

5.5.1 Participant Demographics and Vaccine History

5.5.1.1 Enrollment questionnaire administration

Research staff will administer the enrollment questionnaire or participants may complete electronically. Data will be recorded electronically directly into a data management system such as REDCap.

5.5.1.2 Enrollment questionnaire content

Each participant/participant's LAR(s) will be interviewed to collect information on patient demographics, influenza and COVID-19 vaccination history, and self-reported lab-confirmed influenza or COVID-19 infection in the previous 90 days. Any modifications or additional questions will be submitted for IRB approval before inclusion on enrollment questionnaires. Participant height and weight information may be collected at one of the first three study visits.

5.5.2 Follow-up Surveillance for Vaccine Reactogenicity and Safety and Self-Reported ARI or Change in Health Status Resulting in Hospitalization

5.5.2.1 Follow-up Survey (Reactogenicity)

Participants will complete a brief survey regarding vaccine reactogenicity and safety data 3 days after receipt of either vaccine dose administered according to study procedure (day 0 [groups i-iii] and day 28 [groups ii and iii]).

Follow-up surveys will be completed online. Participants will receive an automated link to an electronic survey 3 days after receipt of either vaccine dose administered according to study procedure (day 0 [groups i-iii] and day 28 [groups ii and iii]) via their preferred contact method (text or email). Participants will complete the survey a maximum of two times during the study period. The first survey will be sent 3 days after Visit 1 (groups i-iii) and the second survey will be sent 3 days after Visit 2 (groups ii and iii). If the survey has not been completed, up to 3 reminder notifications will be sent to the preferred contact method each week.

5.5.2.2 Follow-up Survey (ARI)

Participants will complete a brief survey regarding absence/presence of new ARI symptoms, illness test results (if applicable) and change in health status requiring hospitalization each week.

Weekly surveys will be completed online. Participants will receive an automated link to an electronic survey each week via their preferred contact method (text or email). Participants will complete the survey once per week. The first survey will be sent 7 days after Visit 1 (day 0) and will be sent out each week until the end of the study period (Visit 4; day 180). If the survey has not been completed, a reminder notification will be sent to the preferred contact method each week.

5.5.2.3 Qualifying ARI Definition

An ARI during follow up is defined as an illness with new onset in the past 7 days and any two of the following symptoms: cough, fever or feeling feverish/chills, sore throat, runny/stuffy nose, muscle or body aches, headache, fatigue (tiredness), nausea or vomiting, diarrhea, new loss of smell or taste, or shortness of breath or difficulty breathing.

5.5.3 Study Product Supply, Storage, and Administration

In order to ensure adherence to study randomization assignment, age-appropriate, licensed quadrivalent cell culture-based influenza vaccines (ccIIV4) and EUA or approved mRNA COVID-19 vaccine will be administered as a study procedure. Vaccines will be stored per manufacturer specifications.

5.5.3.1 Quadrivalent cell culture-based influenza vaccine

Flucelvax Quadrivalent (ccIIV4) will be the designated study products for influenza vaccination. A standard-dose of ccIIV4 contains 15 µg of HA per vaccine virus in a 0.5-mL dose. ccIIV4 is approved for persons aged ≥6 months.

Vaccine lot numbers, dose, and site of vaccine administration will be recorded by research staff. After administration, used study syringes will be disposed of according to standard operating procedure.

Emergency management supplies including epinephrine (1:1000) and trained staff will be available for initial treatment of an allergic reaction, if needed.

5.5.3.2 mRNA COVID-19 vaccine

mRNA COVID-19 vaccine will be administered per FDA and ACIP recommendations. Study sites will work to coordinate the brand of mRNA vaccine administered based on recommendations and availability.

Vaccine lot numbers, dose, and site of vaccine administration will be recorded by research staff. After administration, used study syringes will be disposed of according to standard operating procedure.

Emergency management supplies including epinephrine (1:1000) and trained staff will be available for initial treatment of an allergic reaction, if needed.

5.5.4 Biospecimens Collection & Handling

5.5.4.1 Sera

Blood specimens for sera collection will be collected from participants at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), Acute Visit (<10 days after symptom onset), and Convalescent Visit (28 days after Acute Visit). Blood samples will be processed for sera, aliquoted, and stored at -20 C or colder at VE network laboratories until shipped to CDC for storage and subsequent testing via immunological assays to be performed at CDC and/or CDC-approved laboratory facilities (Laboratory Appendix B). We will collect 5-10 mL of whole blood for children aged 6-11 years and 10 mL of whole blood for adults aged 18-64 years. These volumes are preferred for sera processing, however, smaller volumes can be accepted if the situation does not allow for the collection of these amounts. The minimum accepted volume of whole blood to collect is 3 mL for all participants over the age of 6 years. Guidelines will be followed to ensure the amount drawn meets minimal risk criteria (i.e., 3 ml/kg in any 24-hour period, and 7 ml/kg in any eight-week period).

5.5.4.2 PBMC and Plasma

Blood specimens for PBMC and plasma collection will be collected from a subset of participants (target n=250; 200 adults and 50 children) at Visit 1 (day 0), Visit 2 (day 28), Visit 3 (day 56), Visit 4 (day 180), Acute Visit (<10 days after symptom onset), and Convalescent Visit (28 days after Acute Visit). Blood samples will be processed for PBMC and plasma, aliquoted, and stored at VE network laboratories until shipped to CDC for immunologic assays (Laboratory Appendix C). We will collect 8 mL (approximately 1 CPT tube) whole blood for children aged 6-11 years and 24 mL (approximately 3 CPT tubes) whole blood for adults aged 18-64 for PBMC and plasma processing. These volumes are preferred for PBMC and plasma processing,

however, smaller volumes can be accepted if the situation does not allow for the collection of these amounts. The minimum accepted volume of whole blood to collect for PBMC and plasma processing is 16 mL from adults between the ages of 18-64 and 5 mL for children between the ages of 6-11 years. Guidelines will be followed to ensure the amount drawn meets minimal risk criteria (i.e., 3 ml/kg in any 24-hour period, and 7 ml/kg in any eight-week period).

5.5.4.3 Nasal/Throat Swab

Sites may conduct respiratory testing for those that report ARI via the follow-up survey (ARI) during the study period via standard point of care respiratory testing for influenza and COVID-19.

5.6 Vaccine Safety and Reactogenicity

Participants will respond to an automated electronic survey 3 days following each vaccination visit to report the occurrence of fever, injection site reactions (pain, redness, swelling and itching) systemic reactions (chills headache, joint pain, muscle ache, body ache, fatigue, tiredness, nausea, vomiting, diarrhea, abdominal pain, rash lymph node swelling) the severity of reactions and their impact on health. Severity will be rated as mild (symptoms noticeable but aren't a problem) moderate (symptoms limit normal daily activities) or severe (symptoms make normal daily activities difficult or impossible). Impact on health will be assessed with respect to ability of the participant to attend work or school, ability to do normal daily activities and utilization of medical care.

The weekly automatic electronic survey will also request participants to report the occurrence of any change in health requiring hospitalization.

5.7 Medical Record Extraction

5.7.1 Prior Vaccination

All study sites will provide lot number and manufacturer information for documented influenza and COVID-19 vaccination, if recorded, from July 1, 2020 through the end of the study period. The following data elements for vaccination verification will be collected for each dose if provided: date of vaccination, product name, manufacturer, lot number, and source of information, as well as route of administration (e.g., intramuscular, intranasal), valence (e.g., quadrivalent) and type of vaccine (e.g., high-dose, recombinant, standard dose), if applicable. In addition to data sources described below, vaccination cards may also be used as a verified source for COVID-19 vaccination.

5.7.1.1 In-system sources/electronic medical records

Each study site will query the health system's electronic medical record system to obtain prior season influenza and COVID-19 vaccination data for all participants regardless of self-reported vaccination status. Prior influenza and COVID-19 vaccination history data may include records from multiple prior seasons from electronic immunization records.

5.7.1.2 External sources

Each site will, at a minimum, query state and/or local immunization information systems to obtain prior influenza and COVID-19 vaccination data for all participants regardless of self-reported vaccination status. Prior influenza and COVID-19 vaccination data may include records from multiple prior seasons from electronic immunization records. Sites may seek vaccine data from other sources including, but not limited to, occupational health records for major employers, retail pharmacies, and insurance plan claims. A medical release of information (MROI) may be required to be obtained from participants to request data from sources outside the study site's medical system.

5.7.2 Prior Lab-Confirmed Illness

All study sites will provide information for documented influenza and COVID-19 infection, if recorded, from January 1, 2020 through the end of the study period. The following data elements for prior infection will be collected for each lab-confirmed test if provided: date of test, type of test, result of test, and source of information.

5.7.2.1 In-system sources/electronic medical records

Each study site will query the health system's electronic medical record system to obtain prior season influenza and COVID-19 infection data for all participants regardless of self-reported prior infection status. Prior influenza and COVID-19 infection history data may include records from multiple prior seasons from electronic health records.

6 HUMAN SUBJECTS PROTECTIONS

6.1 Institutional Review Board Review

Participating study sites will rely on the Duke IRB. The Duke IRB will enter into an IRB Authorization Agreement (IAA) that will include a communication plan with each relying institution prior to study implementation. Relying study sites will submit study protocols and any amendments and associated consent and assent forms and patient-facing instruments and

recruitment materials to their local IRB for administrative review of the study site reliance as per local IRB policy.

6.2 Participant Confidentiality

According to HIPAA, the information collected by the researchers for this study includes protected health information (PHI). We are requesting a HIPAA waiver of authorization to review electronic medical records. The study meets the requirements of the waiver of HIPAA authorization by ensuring that disclosure of protected health information in this study involves no more than minimal risk to the privacy of individuals. The research could not practicably be conducted without the waiver or alteration and the research could not practicably be conducted without access to and use of the protected health information. The data security plan outlines how identifiers will be protected from improper use or disclosure, how identifiers will be destroyed after study completion, and how the protected health information will not be reused for other research.

Consistent with Section 301(d) of the Public Health Service Act, a Certificate of Confidentiality (CoC) applies to this research because this research is funded, conducted, or supported by CDC and the following is true:

1. The activity constitutes biomedical, behavioral, clinical, or other research; and
    i. Individually identifiable (including coded) information or biospecimens will be obtained or used for research purposes or as defined at 45 CFR 46.102(e);
    ii. Biospecimens are collected or used as part of the research, and is there a small risk that some combination of the biospecimen and other available data sources could be used to deduce the identity of an individual; and
    iii. The research involves information about an individual for which there is at least a very small risk, that some combination of the information, a request for the information, and other available data sources could be used to deduce the identity of an individual.

Therefore, CDC and any of its collaborators, contractors, grantees, investigators or collaborating institutions that receive “identifiable, sensitive information” as defined by subsection 301(d) of the Public Health Service Act shall not:

- Disclose or provide, in any Federal, State, or local civil, criminal, administrative, legislative, or other proceeding “identifiable, sensitive information” that was created or compiled for purposes of the research, unless such disclosure or use is made with the consent of the individual to whom the information, document, or biospecimen pertains; or
- Disclose “identifiable, sensitive information” or provide ISI to any other person not connected with the research.

- Disclosure is permitted only when:
- Required by Federal, State, or local laws (e.g., as required by the Food, Drug and Cosmetic Act or required by state laws requiring the reporting of communicable diseases to State and local health departments), excluding instances of disclosure in any Federal, State, or local civil, criminal, administrative, legislative, or other proceeding;
- Made with the consent of the individual to whom the information, document, or biospecimen pertains; or
- Made for the purposes of other scientific research that is in compliance with applicable Federal regulations governing the protection of human participants in research.

CDC and its collaborators and contractors conducting this research will establish and maintain effective internal controls (e.g., policies and procedures) that provide reasonable assurance that the research is managed in compliance with subsection 301(d) of the Public Health Service Act. CDC will ensure: 1) that any investigator or institution not funded by CDC who receives a copy of identifiable, sensitive information protected by this Certificate, understands that it is also subject to the requirements of the Certificate; and 2) that any subrecipient that receives CDC funds to carry out part of this research involving a copy of identifiable, sensitive information protected by a Certificate understands that it is subject to subsection 301(d) of the PHS Act. Therefore, all study staff will receive training on the importance of protecting the confidentiality of human research participants and of personal information acquired, including the collection of biological specimens.

All research participants will be informed of the protections and the limits to protections provided by this Certificate through the informed consent process. All study staff who obtain consent from study participants will be trained on how the Certificate protects the information collected and the limitations of the Certificate's protections.

6.3 Risks and Benefits to Participants

A description of this clinical trial will be available on http://ClinicalTrials.gov, as required by US Law. This website will not include information that can identify subjects.

The risks to participants for being in this study will be minimal. Getting respiratory samples may cause some discomfort. There may be brief soreness while the nose and throat swabs are taken. The nose swab may cause the participant to sneeze, and once in a while, a small nosebleed could occur. If this should happen, study staff will treat it right away. The participant may experience discomfort or slight pain during blood draw. In rare cases, blood draw may cause bruising, prolonged bleeding, or infection at the site of the puncture.

Some people get severe pain in the shoulder and have difficulty moving the arm where a shot was given. This happens very rarely. Syncope (fainting) can occur in association with

administration of injectable vaccines. Sitting or lying down when space is available for about 15 minutes can help prevent fainting, and injuries caused by a fall, as recommended in the ACIP General Recommendations on Immunization. Participants should inform their doctor if they feel dizzy or have vision changes or ringing in the ears.

As with any licensed or authorized vaccine, protection may not occur in 100% of vaccinated persons for either COVID-19 or influenza vaccines. Participants may be at risk of delayed vaccination if randomized to sequential vaccination groups (groups ii and iii). These participants may be exposed to SARS-CoV-2 or influenza virus before fully immunized. Second vaccine may be offered before visit 2 if risk of COVID-19 or influenza is high based on local viral surveillance.

mRNA COVID-19 Vaccine

Two COVID-19 vaccine received Emergency Use Authorization (EUA) by the FDA in December 2020. BNT162b2 received FDA approval for individuals 16 years of age and older and mRNA-1273 received authorization for those 18 years of age and older. The Pfizer/BioNTech mRNA COVID-19 vaccine received additional EUA for adolescents 12 to 15 years in May 2021 and children 5 to 11 years in October 2021; both the Pfizer/BioNTech and Moderna mRNA COVID-19 vaccines received addition EUA for children aged down to 6 months in June 2022. The EUA was again updated to authorize the use of the updated bivalent vaccines as boosters for children aged down to 6 months in December 2022. These vaccines have been recommended at these ages by the ACIP.

Side effects that have been reported with mRNA COVID-19 vaccines include both injections site reactions as well as general side effects. Injection site reactions include: pain, tenderness and swelling of the lymph nodes in the same arm of the injection, swelling (hardness), and redness at the injection site. General side effects include: fatigue, headache, muscle pain, joint pain, chills, nausea and vomiting, fever and feeling unwell. There is a remote chance that an mRNA COVID-19 vaccine could cause a severe allergic reaction, usually occurring within a few minutes to hours after getting a dose of vaccine. Signs of a severe allergic reaction can include: difficulty breathing, facial and throat swelling, tachycardia, total body rash, dizziness and weakness. There have been rare reports of cases of inflammation of the heart—called myocarditis and pericarditis—happening after mRNA COVID-19 vaccination. The events have mainly occurred in adolescents and young adults and more often after the second dose of vaccine. Available data from short-term follow-up suggest that most individuals have had resolution of symptoms, but information is not yet available about potential long-term sequelae. These may not be all the possible side effects of the mRNA COVID-19 vaccines. Serious and unexpected side effects may also occur. mRNA COVID-19 vaccines are still being studied in clinical trials. Available data support the safety of dose 3 mRNA COVID-19 vaccine in immunocompromised individuals. Available data support the safety of booster doses of mRNA COVID-19 vaccine.

ccIIV4 Influenza Vaccine

ccIIV4 is an FDA-licensed vaccine approved for use in persons 6 months of age and older. This vaccine is standard clinical practice and recommended by the CDC.

ccIIV4 risks include minor problems such as soreness, redness, swelling, or pain where the shot was given, hoarseness, sore, red or itchy eyes, cough, fever, aches, headache, itching, fatigue, all of which usually occur within 1-2 days of vaccination and are self-limiting. More serious problems including a small increased risk of Guillain-Barré Syndrome estimated at 1 or 2 additional cases per million people vaccinated can occur. This is much lower than the risk of severe complications from influenza infection, which can be prevented by ccIIV4. In addition, any medication can cause a severe allergic reaction, or anaphylaxis, which is estimated at ~ 1 in one million doses of ccIIV4 administered.

Allowing Twilio to be Used for Text Message Reminders

We use Twilio to send text messages to participants. Text messaging does not provide a completely secure and confidential means of communication, and the messages are unencrypted. Twilio does encrypt participant information on their servers, but no system is completely safe. If they decide to share these data, it may no longer be covered under the privacy protections. Information that identifies participants, such as phone number, may be sent to and permanently kept by Twilio and their business associates. Information disclosed to these companies or their business partners, may no longer be covered under the privacy protections. Because text messaging does not provide a completely secure and confidential means of communication, a participant may wish to keep communication completely private and we will communicate with the participant only through regular channels like the telephone or email.

Loss of Privacy

There is also the potential risk of loss of confidentiality. Every effort will be made to keep participant's information confidential, however, this cannot be guaranteed.

6.4 Return of Results

Enrolled participants will be informed of any result of FDA-approved laboratory tests used for study purposes. Results of serologic and cellular immunity testing performed as part of this study will not be returned to enrolled patients. Reporting of research test results to public health authorities will adhere to reporting requirements as described in participant terms of consent, and public health authorities may follow up with research participants for further actions per appropriate guidelines.

Reporting of SARS-CoV-2 Results

State or local health department regulations may require reporting of incident cases of SARS-CoV-2 infection. Local investigators at each study site will be responsible for contacting their state or local SARS-CoV-2 surveillance coordinators to ensure study procedures comply with all reporting requirements.

All participants will be informed of positive results once they are available. Results will not be shared with participants' medical providers.

Molecular diagnostic results will not always be available during the period when participants are acutely ill. Participants will be informed via a COVID Test Results Disclaimer.

6.5 Compensation

Participants will be compensated for their participation in the study. Study sites will specify compensation amount and schedule in written/electronic informed consent form .

7 DATA MANAGEMENT, TRANSFER, AND SECURITY

7.1 Data Management and Transfer

Enrollment interview and follow-up survey data will be collected and entered electronically at the sites via Duke's instance of REDCap and stored behind secure institutional firewalls. Linkage to Personally Identifying Information (PII), including names, medical record numbers, postal addresses, phone numbers, email addresses, or other identifying information protected by HIPAA will be securely stored by study sites. Duke will receive names, phone numbers, email addresses, visit dates, and other necessary identifiers as collected through REDCap. A HIPAA-limited dataset will be delivered to CDC by the Network coordinating center (NCC) at Duke University using a unique study-specific identifier for each individual (ENROLLID).

Laboratory data will be linked the unique study identifier (ENROLLID) and Specimen ID to each participant's record and will be uploaded into REDCap by laboratory staff at the central/regional lab. In some sites, study site staff may assist with this task if the laboratory is on-site; other laboratories off-site may directly upload the data into REDCap with minimal interaction with study site staff.

To ensure uniform and consistent data collection among network sites, CDC and/or the NCC will provide data dictionaries and data collection forms/surveys. Data dictionaries will be created based on agreed-upon data elements to be collected through enrollment interview,

follow-up survey, and electronic medical records. Modification to data collection instruments and addition of data requested will be submitted for IRB approval before collection.

All sites are expected to use secure, encrypted file transfer protocols (FTP) or other secure file transfer (upload, download, etc.) to CDC and/or the NCC for data not uploaded directly to the NDCC instance of REDCap. Data will be downloaded by CDC and/or the NCC on a regular basis for quality control checks, tracking, and preparation and cleaning of the final complete dataset. Quality control would include checking data for adherence to the common protocol, outliers, and missing or incomplete data.

7.2 Data Security

Sites will comply with federal, state, and institutional requirements regarding time horizons for retention and/or destruction of research records. Data privacy will be maintained according to local IRB protocols and requirements. No research participant will be identified by name, picture, or any other personally identifying manner if information from this study is presented publicly or published in a medical journal.

Any data stored by each study site prior to upload into REDCap (e.g., medical record and lab results data) will be stored on each site's own servers. Each site must have security policies that comply with all appropriate security requirements. Once data has been uploaded into the NDCC instance of REDCap, it is housed on NCC servers at Duke University. Forms, lists, logbooks, appointment books, and any other listings that link ENROLLID numbers to other identifying information must be stored in a separate, locked (or encrypted) file in an area with limited access. If participant names and corresponding ENROLLIDs are entered into a computer database, this database must be password protected and must be maintained in a directory and in a location that is separate from any study-specific data. The purpose of this requirement is to assure that a breach at one location will not allow the PHI to be associated with other study data.

Deidentified data from this study must be available for release for public use within a year after the data are evaluated for quality as stipulated in CDC Policy 385 ("Policy on Public Health Research and Nonresearch Data Management and Access"). Data that cannot be released publicly (i.e., where deidentification would preclude the proposed analysis) may be shared through a special data-use agreement in accordance with this policy. Data and associated documentation from this study will be available only under a data use agreement developed by CDC in coordination with the NCC that provides for (1) a commitment to using the data only for research purposes and not to identify any individual participant; (2) a commitment to securing the data using appropriate information technology; and (3) commitments for destruction, return, or retention of data as stipulated.

8 STATISTICAL ANALYSIS

8.1 Study Populations

There will be two study populations: the modified Intent-to-Treat (mITT) and Immunogenicity populations. The mITT population will include any participant who was enrolled, randomized, and received at least one vaccine. The Immunogenicity Population is a subset of the mITT Population that includes only subjects who received at least one vaccine, provide visit 1 and visit 2 blood draws available for analysis within the protocol-defined time frame, and with no protocol violations affecting immunogenicity. These protocol violations will be listed in the Statistical Analysis Plan. Statistical analyses of safety outcomes will be performed for the mITT population, and immunogenic outcomes will be analyzed for the Immunogenicity Population.

8.2 Analysis Plan of Year 2 Component D

HAI antibody titers will be compared between groups receiving the mRNA COVID-19 and ccIIV4 influenza vaccines concomitantly or sequentially for each of the four influenza vaccine strains contained in the respective vaccines for that season as well as circulating influenza viruses and SARS-CoV-2 variants. Specifically, we will assess the following objectives:

Primary Objective 1a: Compare the proportion of participants in each vaccination group with a seroprotective HAI titer (≥1:40) pre- and post-ccIIV4 immunization for each ccIIV4 antigen. These proportions will be compared between vaccination groups using an exact Mantel-Haenszel statistic.

Primary Objective 1b: Compare the proportion of participants in each vaccination group achieving seroconversion following ccIIV4 (an HAI titer ≥1:40 following ccIIV4 immunization if the baseline titer is <1:10 or a four-fold rise in HAI titer if the baseline titer is >1:10) for each ccIIV4 antigen. These proportions will be compared between vaccination groups using an exact Mantel-Haenszel statistic.

Primary Objective 1c: Compare the GMT for each ccIIV4 antigen and circulating influenza viruses pre- and post-ccIIV4 immunization in each vaccination group. GMT will be compared between vaccination groups using a regression model.

Primary Objective 1d: Compare the GMFR in HAI titer for each vaccination group. GMFR will be compared between vaccination groups using a mixed effects regression model with a random intercept.

Exploratory objectives will be assessed using exact Mantel-Haenszel statistics to compare proportions between vaccination groups, regression models to compare GMT, and mixed effects regression models to compare GMFR. Tables will be provided summarizing counts and proportions for categorical outcomes or summary statistics and confidence intervals for

continuous outcomes. The statistical analysis of each exploratory objective will be fully described in the statistical analysis plan (SAP).

HAI titers will be log transformed prior to analysis and back transformed to estimate GMT and GMFR. The same analysis plan for HAI titers will be repeated for MN titers.

Data analysis will be performed using SAS version 9.4 software (SAS Institute Inc., Cary, NC) or R software (R Core Team, Vienna, Austria).

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LAB APPENDICES

Lab Appendix A: Respiratory Specimen Testing for Influenza

Lab Appendix B: Human Sera Collection Guidelines for Influenza Serology

Lab Appendix C: Peripheral Blood Mononuclear Cells