Study Protocol

Sneller MC, Liang CJ, Marques AR, et al. A longitudinal study of COVID-19 sequelae and immunity: baseline findings. Ann Intern Med. 24 May 2022. [Epub ahead of print]. doi:10.7326/M21-4905

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Abbreviated Title: COVID-19 Survivor Study

Protocol #: 20I0123

Version Date: December 31, 2020

Title: A Longitudinal Study of COVID-19 Sequelae and Immunity

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STATEMENT OF COMPLIANCE

The protocol will be carried out in accordance with International Conference on Harmonization Good Clinical Practice (ICH GCP) and the following:

- United States (US) Code of Federal Regulations (CFR) applicable to clinical studies (45 CFR Part 46, 21 CFR Part 50, 21 CFR Part 56, 21 CFR Part 312, and/or 21 CFR Part 812)

National Institutes of Health (NIH)-funded investigators and trial site staff who are responsible for the conduct, management, or oversight of NIH-funded trials have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the Institutional Review Board (IRB) for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. In addition, all changes to the consent form will be IRB-approved; a determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.

1 PROTOCOL SUMMARY

1.1 SYNOPSIS

Title: A Longitudinal Study of COVID-19 Sequelae and Immunity

Study Description: This is a longitudinal cohort study to evaluate the clinical sequelae of acute COVID-19 and characterize the immune response to SARS-CoV-2. Household contacts of the COVID-19 cohort will also be recruited and serve as a control group.

Objectives:

- Characterize the medical sequalae and persistent symptoms following recovery from COVID-19 in a cohort of disease survivors.
- Estimate the incidence and risk factors for post-COVID-19 medical sequalae.
- Characterize antibody and cell-mediated immune responses to SARS-CoV-2 in disease survivors
- Characterize evolution of the antibody and T cell-mediated responses to SARS-CoV-2 in survivors over time
- Evaluate survivors for evidence of re-infection with future waves of COVID-19 to determine if initial infection confers long-term protective immunity
- Determine the incidence of clinically silent infection in household contacts
- Characterize the mental health status of survivors and controls including medical trauma related sequelae

Endpoint:

- Establish a clinically well characterized cohort of persons recovered from COVID-19 and close contacts of persons with COVID-19

Study Population: Adult men and women recovering from COVID-19 (n= up to 300) and their household contacts (n= up to 400)

Description of Sites/Facilities Enrolling Participants: National Institutes of Health Clinical Center

Study Duration: 5 years

Participant Duration: 3 years

1.2 SCHEDULE OF ACTIVITIES (SOA)[a]

Procedures	Screening Day-21 to -1	Enrollment/Baseline Mo. 0[b]	Study Visit-2 Mo. 6[c]	Study Visit-3 Mo. 12[c]	Study Visit 4 Mo. 18[c]	Study Visit 5 Mo. 24[c]	Study Visit 6 Mo. 30[c]	Final Study Visit Mo. 36[c]
Informed consent	X							
SARS-CoV-2 RNA testing	X							
Medical history		X	X	X	X	X	X	X
Physical exam		X	X	X	X	X	X	X
Vital signs		X	X	X	X	X	X	X
Height		X						
Weight		X	X	X	X	X	X	X
Study questionnaire		X	X	X	X	X	X	X
Hematology		X	X	X	X	X	X	X
Plasma chemistry [d]		X	X	X	X	X	X	X
Urinalysis		X						
Pregnancy test [e]		X	X	X	X	X	X	X
HIV, HBV, and HCV serology; antiphospholipid antibody; quantitative immunoglobulins		X						
HLA typing		X						
Peripheral blood lymphocyte flow cytometry		X		X		X		X
Pulmonary function testing[f]		X[j]		X		X		X
Electrocardiogram (ECG), echocardiogram		X[j]						
Blood samples for research/storage (e.g., immunology assays)		X	X	X	X	X	X	X
Optional leukapheresis[g]		X[i]	X	X	X	X	X	X

Procedures	Screening Day-21 to -1	Enrollment/Baseline Mo. 0[b]	Study Visit-2 Mo. 6[c]	Study Visit-3 Mo. 12[c]	Study Visit 4 Mo. 18[c]	Study Visit 5 Mo. 24[c]	Study Visit 6 Mo. 30[c]	Final Study Visit Mo. 36[c]
Chest Radiograph[h]		X						
Mental health evaluation[k]		X	X	X	X	X	X	X

a: additional visits may occur for urgent evaluation of symptoms possibly due to recurrent COVID-19. Participants who receive a SARS-CoV-2 vaccine from their employer or private physician may have an optional blood draw visit 5-14 days following vaccination.
b: within 14 days of screening visit
c: visit window +/- 6 weeks for all visits
d: albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, phosphorus, potassium, total protein, AST, ALT, sodium, CRP, troponin, pBNP, rheumatoid factor, antinuclear antibody
e: pregnancy test (women of childbearing potential) done prior to each leukapheresis.
f: spirometry, lung volumes, DLCO and 6-minute walk test; for control/contact cohort, testing done only at baseline or 6-month visit
g: may occur anytime during visit window
h: to be done at baseline on COVID-19 cohort only
i: Baseline Apheresis to be done within 28 days of Enrollment
j. Baseline PFTs/6MW, echocardiogram, electrocardiogram (ECG) to be done within 2 months of Enrollment
k. Baseline mental health interview and surveys; online surveys every 6 months; participant interview yearly.

2 INTRODUCTION

2.1 STUDY RATIONALE

Coronavirus disease-2019 (COVID-19) is a newly recognized viral disease primarily involving the respiratory tract. The world is currently experiencing COVID-19 pandemic with a significant portion of those infected developing severe disease manifestations including pneumonia, acute respiratory distress syndrome with respiratory failure, and, in some cases, death. The clinical characteristics and spectrum of disease severity of acute COVID-19 are currently being defined [1-4]. Since COVID-19 is a new disease, very little is known about possible clinical sequelae that may persist after resolution of the acute infection. In addition, the characteristics of the initial cellular immune and antibody response to SARS-CoV-2 (the virus that causes COVID-19) have not been fully defined and it is not known if the immune responses generated by infection provides long-term protective immunity. The purpose of this study is to establish a longitudinal cohort that has recovered from COVID-19 and characterize the clinical sequelae of acute infection, characterize the immune response to the virus, and follow the evolution of the immune response over time and determine the extent to which natural immunity is protective against re-infection.

2.2 BACKGROUND

SARS-Co-V-2 is a novel coronavirus not previously known to infect humans. SARS-CoV-2 predominantly infects the upper and lower respiratory tract and causes a spectrum of clinical manifestations ranging from mild upper respiratory tract symptoms to severe pneumonia with respiratory failure [1-5]. The clinical syndrome produced by infection with SARS-CoV-2 is referred to as COVID-19. Current evidence suggests that in a significant portion of individuals, infection may be subclinical [5].

SARS-CoV-2 appears to be transmitted via respiratory droplets. Person-to-person transmission occurs when infected individuals emit droplets containing virus particles while coughing, sneezing, or talking. The first COVID-19 cases were reported in December 2019 in Wuhan, China. Infection with SARS-CoV-2 quickly spread first within Wuhan and subsequently to other regions of China and the world. By early March, over 100,000 cases of COVID-19 were reported in 114 countries. On March 11, 2020 the WHO made the assessment that COVID-19 can be characterized as a pandemic. The first case in the United States was identified in Washington state on January 20, 2020. By the first week in April, over 230,000 cases of COVID-19 had been reported in the United States [6].

Rates of hospitalization and case fatality vary in different countries. In the United States, the overall rate of COVID-19 hospitalizations was 12.3 per 100,000 population with a highest rate in those 65 years of 38.7 per 100,000 [7]. As of April 11, 2020, in Maryland, 22% of documented COVID-19 cases require hospitalization (Maryland Department of Health Web site). The case fatality rate is difficult to determine given the uncertainty regarding the total number of cases. Based on current reporting to the Centers of Disease Control, the case fatality rate in the United states is 3.5% with 80% of deaths occurring in those 65 years and older [7]. However, because of the uncertainty in the denominator, this is not a reliable estimate of the case fatality rate.

Given the recent onset of the COVID-19 epidemic, the long-term sequelae of infection are unknown. Limited information of disease sequelae is available from the SARS-1 coronavirus outbreak in 2003. In a study of 110 survivors of 2003 SARS-1 outbreak in Hong Kong, significant impairment in pulmonary function was noted in 15% of survivors 6 months after recovery. In addition, exercise capacity and health status of these SARS-1 survivors were considerably lower than that observed in the general population [8]. Due the frequency of pneumonia in COVID-19, long term pulmonary sequelae are likely. Non-pulmonary clinical manifestations have also been described in hospitalized patients with COVID-19 [3,4,9] and, in some cases, after recovery[10] thus, long-term sequelae in other organ systems may manifest after resolution of the acute disease.

Information regarding the immune response to SARS-CoV-2 infection is just beginning to emerge. Based on data from China, IgM antibody response is detected a median of 5 days after symptom onset, with IgG responses appearing at a median of 14 days [11]. There is currently no data regarding the decline in anti-SARS-CoV-2 antibodies over time. Data from past SARS-CoV-1 and Coronavirus 229E infections indicate that titers of IgG antibodies decline to low levels 1-3 years after infection [12,13]. These findings indicate that the antibody response to infection with other known coronaviruses wanes over 1-3 years. If immunity to SARS-CoV-2 is short-term, this would favor the establishment of annual or biennial outbreaks of COVID-19[14]. As the COVID-19 pandemic is only a few months old, there is no data on the decline in antibody levels with time; thus, it is not known how long any protective immunity from natural SARS-CoV-2 infection might last.

This study will establish a cohort of COVID-19 survivors to study the long-term sequelae of acute infection and the evolution of the immune response over time. Longitudinal follow-up of this cohort will provide important information about clinical sequelae of acute COVID-19, characteristics of the immune response to SARS-CoV-2, and the extent and duration of protective immunity.

In addition to the respiratory and other organ system involvement caused by SARS-CoV-2, most experts agree that there is a significant mental health toll due to restrictions of normative social behaviors and isolation, e.g. social distancing[15]. The environmental stressors imposed by constraints on activities, social contact, and access to resources are significant and are experienced by most Americans. Furthermore, there have been reports of adverse mental health sequelae from exposure to traumatic events by health care workers who cared for COVID-19 patients during their acute infection or hospitalization [16]. Medical trauma to survivors can result from medical procedures, illnesses, and hospital stays and can lead to clinically significant conditions such as posttraumatic stress disorder, anxiety and mood disorders, as well as persistent somatic complaints[17]. It is not known what the rehabilitative, cognitive, and mental health needs of COVID-19 survivors will be once they recover from their acute illnesses[18].

2.3 RISK/BENEFIT ASSESSMENT

2.3.1 Known Potential Risks

Risks of Phlebotomy

Phlebotomy may be associated with discomfort, bruising, local hematoma formation and, on rare occasions, infections, lightheadedness, and fainting. The amount of blood drawn for research purposes will be within the limits allowed for adult participants by the National Institutes of Health (NIH) CC (Medical Administrative Policy 95-9: Guidelines for Limits of Blood Drawn for Research Purposes in the Clinical Center: http://cc-internal.cc.nih.gov/policies/PDF/M95-9.pdf).

Risks of mental health evaluation

The mental health evaluation will involve yearly interviews and surveys of topics, such as trauma exposure, anxiety and mood symptoms. The risks associated with clinical interviews are minimal, beyond transient increases in stress associated with difficult topics. The interviews will be conducted by a trained mental health clinician who can follow-up on clinically significant issues. It is possible a participant may report high levels of mental health symptoms, onset of new symptoms or show trends in worsening of symptoms that could indicate a need for mental health care. If there is a question of participant safety, or a harm to others e.g. suicidal ideation, a consultation to the NIMH Psychiatric Consultation Liaison Service will be requested. At a minimum, we will provide information about national and local mental health resources, such as hotlines, and guidance on steps to take to seek care in the community for any individual who could benefit.

The interview may be audio-recorded to facilitate qualitative analysis. If interviews are recorded, the participant will sign NIH Form 661 that authorizes recording of patients for research purposes. The participant can decline recording. Audio recording carries the risk that the participants could be identified. All recordings will be digital and identified by study number without other identifiers. These recordings will be stored on secure servers that are password protected and only members of the research team will have access to them. Recordings will be destroyed when the research is complete.

Risks of Leukapheresis

The potential risks associated with leukapheresis include lightheadedness, dizziness, possible fainting, tingling around the mouth and in the fingers and toes, nausea, chills, vomiting, mild muscle cramps, loss of <1 pint of blood, or pain, bruising, or discomfort at the needle insertion sites. More serious, but rare, complications include nerve damage at the needle insertion site, seizures and air embolism. Most procedures are performed without an incident. Blood components removed during leukapheresis are generally replaced by the body within a few hours or a few days. No infections associated with this procedure have been reported in thousands of cases performed over the last 10 years at the NIH.

Risks of a Single Anterior-Posterior (AP) Chest Radiograph

A single standard AP chest radiograph delivers 0.1mSv of radiation which is comparable to 10 days of natural background radiation.

Risks of Pulmonary Function Testing (PFT)

Pulmonary function testing is not an invasive procedure. Some patients with underlying lung disease may experience wheezing, coughing, shortness of breath, chest tightness, dizziness, and headache during the test. If this happens, we will stop the test until symptoms resolve.

Risks of HLA Typing

Some human leukocyte antigen (HLA) types have been associated with an increased risk of certain diseases like arthritis and other rheumatologic disorders, or a faster progression to acquired immunodeficiency syndrome (AIDS). HLA typing will be performed on samples collected from all the enrolled participants. Results from the HLA typing will become part of each participant's medical record at NIH. Medical records containing this information are maintained in a secure place.

Risks of Electrocardiogram (ECG) and Echocardiogram

There are no risks of harm associated with these two procedures.

2.3.2 Known Potential Benefits

There are no direct benefits to the patient for participating in this study. Knowledge gained from this study may provide important new information on the clinical sequalae of COVID-19 and the immune response to the virus. Such knowledge could produce benefits for society at large.

2.3.3 Assessment of Potential Risks and Benefits

The potential risks of participation in this study are minimal and balanced by the potential of this study to provide important new knowledge about sequalae of COVID and the development of protective immunity following natural infection with SARS-CoV-2.

3 OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS
Primary		
• Characterize the medical sequelae and persistent symptoms following recovery from COVID-19 in a cohort of disease survivors.	• Establish a clinically well characterized cohort of persons recovered from COVID-19 and close contacts of persons with COVID-19 • Establish a clinically well characterized cohort of persons	COVID-19 is a new disease and establishing this cohort is the best way to meet the study objectives

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS
• Determine the incidence and risk factors for post-COVID-19 medical sequelae. • Characterize antibody and cell-mediated immune responses to SARS-CoV-2 in disease survivors • Characterize evolution of the antibody and T cell-mediated responses to SARS-CoV-2 in survivors over time • Evaluate survivors for evidence of re-infection with future waves of COVID-19 to determine if initial infection confers long-term protective immunity • Characterize the mental health status of survivors and controls including medical trauma related sequelae • Determine the incidence of clinically silent infection in close contacts	recovered from COVID-19 and close contacts of persons with COVID-19 who are willing to donate blood for the research studies outlined in Section 6.3.1	COVID-19 is a new disease and establishing this cohort is the best way to meet the study objectives
Secondary		
N/A	*N/A.*	*N/A*
Tertiary/Exploratory		
N/A	*N/A.*	*N/A*

4 STUDY DESIGN

This is an observational cohort study of individuals who have recovered from COVID-19. Participants will be evaluated at National Institutes of Health (NIH) Clinical Center (CC). For consenting participants who meet the eligibility criteria, a medical history will be obtained, a physical examination will be performed, and blood samples will be collected.

Demographic information and details of acute COVID-19 illness (including dates of hospitalization), for COVID-19 obtained from participants and any available medical records.

Study participants will be followed for 3 years.

At the baseline examination, COVID-19 survivors will be asked about household members since the time of the diagnosis with COVID-19. Household members will be invited to enroll in the study as a control cohort. For consenting household contact participants, a medical history will be obtained, a physical examination will be performed, and blood samples will be collected. Information on contact with the COVID-19 survivor and any self-reported symptoms occurring within 2 weeks of the survivor's illness will be collected.

Both COVID-19 survivors and household contacts will be tested for antibody and T cell-mediated responses to SARS-CoV-2. In the latter group, this will allow us to assess for possible unrecognized SARS-CoV-2 infection.

When data are available on the baseline characteristics of the COVID-19 survivors and the prevalence of medical complications, a decision will be made on the selection of an appropriate control group from the enrolled household contacts. Those without serologic evidence of prior infection with SARS-CoV-2 will be used as controls for comparing the prevalence and incidence of medical complications with COVID-19 survivors.

Other individuals without known COVID-19 (such as exposed healthcare workers) may also be invited to participate in the study. Those individuals, without serologic evidence of prior SARS-CoV-2 infection, may also be used as controls for comparing the prevalence and incidence of medical complications with COVID-19 survivors.

Following the Baseline Visit, all participants will be seen every 6 months for a total of 3 years. Study visits may occur over several days within the study visit window period. Additional visits may occur as clinically indicated in the COVID-19 cohort for evaluation of intercurrent respiratory tract illness. Participants who receive a SARS-CoV-2 vaccine from their outside health provider or employer may have an optional blood draw visit. The purpose of this optional visit is to obtain peripheral blood by venipuncture for the quantitative and qualitative evaluation of early vaccine-induced B-cell response in individuals who were previously infected with SARS-CoV-2 and in uninfected controls using the assays described in Section 7.3.1.

Standard diagnostic studies and consultations will be performed as clinically indicated in accordance with standard medical practice for evaluation of symptoms and abnormal physical findings. No medical treatment for non-emergency medical conditions identified during the study will be provided by the NIH. If medically conditions requiring non-emergency treatment are identified during protocol participation, participants will be referred to their outside physician or clinic for treatment.

4.1 CONCOMITANT THERAPY

N/A

5 STUDY POPULATION

5.1 INCLUSION CRITERIA

In order to be eligible to participate in this study, all individuals must meet all of the following criteria:

1) Stated willingness to comply with all study procedures and availability for the duration of the study

2) Age 18 years or older.

3) Ability of participant to understand and the willingness to sign a written informed consent document.

4) Hemoglobin of 9.0 gm/dl or higher

5) Willingness to give consent for the storage of blood samples for research.

6) Have a physician or clinic outside NIH to manage underlying medical conditions or agreeing to establish care with an outside physician or clinic for any medical conditions requiring treatment that may be diagnosed as a result of protocol participation.

COVID-19 Survivor Group

1) Documented prior COVID-19 as evidenced by:

   a) detection of SARS-CoV-2 RNA or antigen in nasopharyngeal swab, sputum or other sample source with EUA/approval from the FDA; or

   b) a positive antibody test using an assay that has received emergency use authorization (EUA) from the Food and Drug Administration (FDA) and a history clinical manifestation compatible with COVID-19.

2) Greater than 6 weeks since onset of COVID-19 symptoms and no fever for at least 1 week. For individuals with asymptomatic infection, screening will not occur until at least 4 weeks after the last positive SARS-CoV-2 PCR or antigen test.

COVID-19 Close Contact

1) Living in the same household as a COVID-19 survivor during the time of illness or, being within approximately 6 feet (2 meters) of a COVID-19 case for a prolonged period of time or having direct contact with infectious secretions of a COVID-19 case (e.g., being coughed on).

2) No diagnosis of COVID-19 or current symptoms suggestive of COVID-19

5.2 EXCLUSION CRITERIA

An individual who meets any of the following criteria will be excluded from participation in this study:

1. Current abuse of alcohol or other drugs that, in the judgement of the Principal Investigator (PI) could interfere with patient compliance.

2. Inability to travel to the NIH Clinical Center for study visits

3. Any medical or mental health condition that, in the judgement of the PI, would make the volunteer unable to participate in the study.

4. Positive SARS-CoV-2 PCR at screening visit.

5. History of any of the following in the past 14 days: fever > 38.2^{o} C; new or worsening respiratory symptoms (e.g. cough, dyspnea).

6. Pregnancy

5.3 INCLUSION OF VULNERABLE PARTICIPANTS

Pregnant Women will not be enrolled in the protocol. If a woman is pregnant at the time of screening she will not be enrolled until the end of her pregnancy because important baseline research assessments (e.g. leukapheresis, chest x-ray, pulmonary function testing) should not be done during pregnancy.

Children will not be included in this study. Due to the rarity of COVID-19 in children (as of 4/11/2020 in Maryland, <2% of cases occurred in children), it would not be possible to recruit enough children to address the protocol research objectives. In addition, pediatric restrictions for the volume of blood permitted to be drawn for research purposes and the more than minimal risk of apheresis in children would severely limit the ability to conduct the immunologic studies described in Section 7.3.1.

5.3.1 Participation of Employees

NIH employees may be enrolled in this study as this population meets the study entry criteria. Neither participation nor refusal to participate as a participant in the research will have an effect, either beneficial or adverse, on the participant's employment or position at NIH.

Every effort will be made to protect participant information, but such information may be available in medical records and may be available to authorized users outside of the study team in both an identifiable an unidentifiable manner.

The NIH Information Sheet on Employee Research Participation will be made available. Please see section **9.1.5** for consent of employees.

5.4 LIFESTYLE CONSIDERATIONS

N/A

5.5 SCREEN FAILURES

Individuals who do not meet the criteria for participation in this study (screen failure) may be re-screened after a minimum time of 4 weeks.

5.6 STRATEGIES FOR RECRUITMENT AND RETENTION

The target sample size of this study is up to 300 COVID survivors and up to 400 household contacts. Up to 400 COVID survivors and 500 contacts may be screened to reach target enrollment. Given the large numbers of COVID cases in the Baltimore-Washington, DC metropolitan area, sufficient numbers of participants can be recruited from the local area.

Local and regional recruitment will be done using directed mailings to local physicians, internet ad campaigns, social media outlets, print ads, and from local clinics via the NIAID Office of Communications and the NIAID patient recruitment with Matthews Media Group, Inc. Patients participating in local-regional acute COVID -19 treatment trials (such as NIAID 20-0006) may also be recruited after they complete the parent study.

To optimize retention on the study, participants will be compensated for each study visit, with additional bonus compensation for completion of all visits. Participants who live within a 50-mile radius of the NIH CC, will be offered taxi transportation for study visits via the NIAID patient recruitment contract with Matthews Media Group, Inc.

5.6.1 Costs

Study-related evaluations will be provided at the NIH CC at no cost to the participant. Participants will be reimbursed for travel and lodging expenses per the NIAID Travel Policy.

5.6.2 Compensation

Participants will receive financial compensation for time and inconvenience according to the NIH CC volunteer guidelines:

- $100 for the screening visit.
- $50 for individual study visits

- $200 for each leukapheresis procedure (up to 2 per year), or $70 for extra blood if participant does not undergo the apheresis procedure.
- $400 for completion all study visits.

Compensation for NIH employees will be in accordance with OHSRP SOP 14F

6 PARTICIPANT DISCONTINUATION/WITHDRAWAL

6.1 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

- Request by the patient to withdraw from the study
- Refusal to comply with study guidelines or procedures
- Repeatedly missing scheduled appointments
- If a patient is lost to follow-up for more than 8 months
- If a study investigator thinks that staying in the study is not in the patient's best interest
- If a patient does not respect the property of the U.S. Government, other patients, or NIH staff
- If, in the judgment of a study investigator, a patient exhibits inappropriate or threatening behavior towards NIH staff or other patients
- Refusal to allow continued drawing of blood for research sample storage

The reason for participant discontinuation or withdrawal from the study will be recorded on the CRIMSON off study note.

6.2 LOST TO FOLLOW-UP

A participant will be considered lost to follow-up if he or she fails to return for more than 8 months and is unable to be contacted by the study staff.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site will attempt to contact the participant and reschedule the missed visit and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain if the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee will make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record or study file.
- Should the participant continue to be unreachable, he or she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

7 STUDY ASSESSMENTS AND PROCEDURES

7.1 SCREENING PROCEDURES

Procedures described in this section will be done on both COVID-19 survivors and household contact/controls.

7.1.1 Screening activities performed prior to obtaining informed consent

Potential participants will be pre-screened via review of outside medical records to assure they meet the criteria listed in Sections 5.1 and 5.2. Health Insurance Portability and Accountability Act (HIPAA) rules, other relevant federal or state laws, and local institutional requirements will be followed, in obtaining outside medical records for review.

7.1.2 Screening activities performed after a consent for screening has been signed

The following activities will be performed only after the participant has signed the consent this study. Assessments performed at outside facilities or on another NIH protocol within the timeframes below may also be used to determine eligibility once a patient has signed the consent.

The screening process will occur in 2 phases. At the initial screening visit informed consent will be obtained (if not previously obtained by phone) and the participant will have a nasopharyngeal swab (or other sample source with EUA/approval from the FDA) obtained for SARS-CoV-2 polymerase chain reaction (PCR). An optional complete blood count (CBC) may be done at the initial screening visit if research apheresis is scheduled for the second Screening/Enrollment visit. If the SARS-CoV-2 PCR results return negative, the participant will return for a second Screening/Enrollment visit at which the procedures listed below will be performed. This visit will occur within 14 days of the initial Consent-Screening visit. Unless otherwise noted, both participants recovering from COVID-19 and household contacts will undergo identical clinical evaluations.

- Medical history and physical examination, including weight and vital signs.

- Mental health evaluation may be conducted by licensed mental health care clinicians using a semi-structured interview about illness experiences, trauma exposure and past mental health history including substance use. This may include a diagnostic evaluation of current and past psychiatric disorders. These interviews may take place in person at NIH or via NIH Clinical Center telemedicine service. The interview may be recorded to facilitate data collection and qualitative analysis as described in Section 2.3.1.
- Participants may be asked to complete assessments about cognition (NIH ToolBox) and self-report surveys such as the Patient Health Questionnaire (PHQ 9), WHODAS 2.0, Impact of Events Scale or Adverse Childhood Events (ACEs) and 36-Item Health Survey questionnaire (SF-36), which query mental health symptoms, history and functional status (http://www.healthmeasures.net/explore-measurement-systems/nih-toolbox/intro-to-nih-toolbox/cognition).

- Questionnaires can be filled using a paper packet or entered directly into a research database, Research Electronic Data Capture (REDCap). REDCap is an established electronic data collection tool used at NIH. Additional questionnaires may be added based on new findings from the study.

- Blood collection for:

    - HIV and Hepatitis C antibody serology

    - HBsAg

    - Anti-phospholipid antibodies

    - Quantitative immunoglobulins

    - Complete blood count (CBC) with differential, prothrombin time (PT), activated partial thromboplastin time (PTT), D-dimer

    - Chemistry panel to include ALT, AST, alkaline phosphatase (ALP), creatinine, total and direct bilirubin, serum albumin levels, troponin, pro-brain natriuretic peptide (BNP), c-reactive protein (CRP), rheumatoid factor, antinuclear antibody (ANA)

    - Peripheral blood lymphocyte flow cytometry panel

    - Storage of peripheral blood mononuclear cells (PBMC), serum, and plasma

    - HLA Typing-low resolution (if not already on file)

- Additional standard laboratory test may be added as new information emerges about the clinical spectrum of COVID-19

- Urinalysis

- Optional Leukapheresis

- Serum or urine pregnancy test for women of child-bearing potential prior to leukapheresis

- Standard pulmonary function testing and 6-minute walk testing (For COVID-19 cohort done within 2 months of enrollment; for close contact/control cohort done at either baseline or 6-month visit)

- Electrocardiogram (ECG) and echocardiogram

- Anterior-posterior chest radiograph (COVID-19 cohort only)

No additional cardiac imaging will be performed on this protocol. Participants will be offered co-enrollment in the NHLBI Cardiac MRI Protocol (18-H-0118).

7.2 CLINICAL EVALUATIONS

Unless otherwise noted, all evaluations will be done on both COVID-19 survivors and contacts. Refer to table in Section 1.2 for frequency.

- Physical examination medical history and a targeted physical examination, including weight, vital signs, and a symptom-directed evaluation based on symptoms or complaints reported by each participant. History and physical examinations will be done by a Physician, Physician's Assistant, or Nurse Practitioner.

- Mental health evaluation may be conducted by licensed mental health care clinicians using a semi-structured interview about illness experiences, trauma exposure and past mental health history including substance use. This may include a diagnostic evaluation of current and past psychiatric disorders. These evaluations may take place in person or via NIH Clinical Center telemedicine service. The interview may be recorded to facilitate data collection and qualitative analysis as described in Section 6.1.2.

- Participants may be asked to complete assessments about cognition (NIH ToolBox) and self-report surveys such as the Patient Health Questionnaire (PHQ 9), WHODAS 2.0, Impact of Events Scale or Adverse Childhood Events (ACEs) and 36-Item Health Survey questionnaire (SF-36), which query mental health symptoms, history and functional status. Questionnaires can be filled using a paper packet or entered directly into a research database, Research Electronic Data Capture (REDCap). REDCap is an established electronic data collection tool used at NIH. Additional questionnaires may be added based on new findings from the study.

- Standard pulmonary function testing and 6-minute walk testing (done yearly; COVID-19 cohort only)

- Standard diagnostic studies and consultations will be performed as clinically indicated in accordance with standard medical practice for evaluation of symptoms and abnormal physical findings (COVID-19 cohort only). Examples of such testing may include (but are not limited to) chest radiography and/or lung CT scan, electrocardiogram, echocardiogram, and medical subspecialty consultations.

7.3 BIOSPECIMEN EVALUATIONS

At study follow-up visits, the following biospecimens will be obtained from both COVID-19 survivors and household contacts/controls.

- Blood collection for:
    - Complete blood count (CBC) with differential
    - Chemistry panel to include ALT, AST, alkaline phosphatase (ALP), creatinine, total and direct bilirubin, serum albumin levels, troponin, pro-BNP, and CRP
    - Peripheral blood lymphocyte flow cytometry panel (yearly only)
    - Storage of peripheral blood mononuclear cells (PBMC), serum, and plasma for research studies described in 7.3.1
- Optional leukapheresis for research studies described in 7.3.1
- Serum or urine pregnancy test for women of child-bearing potential prior to leukapheresis.
- Additional standard laboratory studies may be performed in accordance with standard medical practice for evaluation of symptoms and abnormal physical findings.
- All biospecimen evaluations will be performed by the either AIDS MONITORING Laboratory, the Virus Isolation and Serology Laboratory, Leidos Biomedical LEIDOS BIOMEDICAL RESEARCH, INC or the NIH CC LABORATORY using commercially available methods. The laboratories are Clinical Laboratory Improvement Amendments (CLIA) certified to perform the above testing.

7.3.1 Correlative Studies for Research

<u>Serologic assay for anti-SARS-CoV-2 antibodies</u>

Assays for serum antibodies will be performed at all visits using commercial assays approved under FDA Emergency Use Authorization (EUA). Additional research antibody tests may also be used to evaluate aspects of the SARS-CoV-2 antibody response including (but not limited to) titers of viral neutralizing antibodies done to be done at the NIAID high-containment laboratory at Ft. Detrick, MD.

<u>B-cell and T-cell and biomarker studies in COVID survivors and household contacts</u>

A full characterization of the antibody response to SARS-CoV2 in people who have recovered from COVID-19 and their household contacts involves assessing serum or plasma antibodies and the B cells that generate and sustain them. In individuals who have recovered from viral infections, memory B cells that are specific for the virus can be detected in the peripheral blood within weeks of infection, with kinetics, potencies, effector function and longevities that remain largely unknown for SARS-CoV2. A variety of approaches will be used to evaluate SARS-CoV2-specific B cells in the peripheral blood of participants enrolled, including flow cytometry, ELISA/ELISPOT, and single-cell approaches with barcoded antibodies against cell-surface markers and viral antigens. Barcoded and fluorophore-label SARS-CoV2 antigens are being designed for this purpose.

An extensive phenotypic and functional characterization of T cell responses to SARS-CoV2 in people who have recovered from COVID-19 and their close contacts is necessary to fully comprehend the impact of viral infection on host effector immune system. These analyses will be conducted by multi-color flow cytometry and multiplexed cytokine analyses of antigen-specific T cells. The composition of T cell subsets in blood of the study participants will be evaluated based on expression of CD45RA, CD27, and CCR7 on $CD3^+CD4^+$ and $CD3^+CD8^+$ T cells.

Frequencies of naive, central memory, effector, transitional memory, and effector memory RA $CD4^+$ and $CD8^+$ T cells in peripheral blood mononuclear cells (PBMCs) of individuals who have recovered from COVID-19 will be monitored over time and compared to those of the age and gender-matched close contacts. In addition, levels of cellular/immune activation, immune exhaustion/inhibitory receptors, and immune costimulatory receptors on each T cell subset will be monitored over time to evaluate the degree and extent of immunologic abnormalities that may persist in COVID survivors and their close contacts.

A large number of biomarkers may be involved in the immune response to COVID-19, including cytokines, chemokines and markers of inflammation. Accordingly, multiplexed proteomic assays capable of detecting 50+ soluble proteins will be performed on plasma and in a single-cell platform on pathogen (SARS-CoV2, tetanus, and CMV)-specific $CD4^+$ T or $CD8^+$ T or B cells from COVID survivors and their close contacts.

To examine the evolution of the memory T-cell response to SARS-CoV-2, PBMC samples from multiple convalescent time points (4-6x10E6 cells per time point) will be stimulated with appropriate control antigens or viral peptide pools spanning conserved structural proteins like the spike (S) protein. Cells will be analyzed by flow cytometry to measure the magnitude of cytokine-secreting, virus-specific $CD4^+$ and $CD8^+$ T-cell responses, frequency of epitope-specific $CD8^+$ T cells by HLA class I/SARS-CoV-2 multimer labeling in individuals bearing a specific HLA type, and phenotypic marker expression denoting the states of maturation (CCR7, CD45RO), activation (Ki67, PD-1) and exhaustion (PD-1). Functionality will be assessed in flow cytometry-based assays by intracellular expression of antiviral cytokines, the CD107a degranulation marker, and the cytotoxic proteins perforin and granzyme B. Exploratory analyses of other functional parameters, including cytotoxicity of peptide-pulsed autologous targets, will be attempted when PBMC numbers from a given time point exceed 6-8x10E6 cells. Comparisons and correlations will be made between clinical outcomes (stratified by disease severity) and flow cytometry results.

The above described assays will be performed longitudinally in real time or retrospectively on cryopreserved plasma and peripheral blood mononuclear cells isolated by density gradient centrifugation (Ficoll) from blood samples obtained from study participants. Additional *in vitro* immunologic studies may be performed based on new information generated during the conduct of this study.

7.3.2 Samples for Genetic/Genomic Analysis

Genetic testing/genomic analysis will not be done under this protocol. All participants will be offered co-enrollment in the NIAID Centralized Sequencing Protocol (17-I-0122).

8 STATISTICAL CONSIDERATIONS

8.1 STATISTICAL HYPOTHESIS

This protocol describes an observational study that will be descriptive in nature. The primary goal is to characterize COVID-19 survivors in terms of their medical sequalae and persistent symptoms, incidence and risk factors for the medical sequalae, and antibody and cell-mediated immune responses to SARS-CoV-2. Recruitment of controls will allow more precise attribution of the aforementioned measurements to COVID-19 than would be possible without a control group.

Data collected from this study may also provide the opportunity to collect evidence of re-infection from SARS-CoV-2 and to learn more about whether an initial infection confers long-term protective immunity. Additionally, data from controls is expected to provide information about the incidence of clinically silent infection among household contacts.

8.2 SAMPLE SIZE DETERMINATION

Based on resource availability, it is expected that the study will be able to enroll up to 300 COVID-19 survivors and 400 controls. For simplicity, sample size and power calculations are made assuming a subset of the same number of controls as there are survivors is selected. This is plausible, in the scenario that approximately equal numbers of household and non-household controls are selected, and separate comparisons are made against the two different control groups. For example, for a binary outcome of interest that has a 5% incidence in the control group, enrolling 200 COVID-19 survivors and comparing them with 200 controls would allow for detecting a relative risk of 2.6 with 80% power at 0.05 significance (2-sided). The below table shows the detectable relative risk at 80% power and 0.05 significance (2-sided) for a variety of sample sizes and incidence rate of outcome in the control group.

Relative risk of outcome (e.g. sequelae, symptom, immunological endpoint) that can be detected with 80% power at 0.05 significance (2-sided) when comparing COVID-19 survivor group to control group					
Expected incidence in control group	N=100 per group	N=200 per group	N=300 per group	N=400 per group	N=500 per group
1%	9.0	5.9	4.7	4.1	3.7
2%	5.8	4.0	3.3	2.9	2.7
5%	3.4	2.6	2.3	2.1	1.9
10%	2.4	2.0	1.8	1.7	1.5
20%	1.9	1.6	1.5	1.4	1.4

Depending on how strongly an initial infection confers protective immunity, COVID-19 survivors in this study may become re-infected during the course of the study. If, for each COVID-19 survivor, there is a 1% chance over the course of 3 years to become re-infected, with 200 COVID-19 survivors, there is an 87% chance that at least one survivor will become re-infected. The table below lists the probability of observing at least one COVID-19 survivor experiencing re-infection, for a range of sample sizes and individual probabilities of re-infection.

Probability that at least one COVID-19 survivor will become re-infected					
Probability of re-infection over a 3-year period for each survivor	N=100 survivors	N=200 survivors	N=300 survivors	N=400 survivors	N=500 survivors
0.25%	22%	39%	53%	64%	71%
0.5%	40%	64%	78%	86%	92%
1%	63%	87%	95%	98%	99%
2%	87%	98%	>99%	>99%	>99%
4%	98%	>99%	>99%	>99%	>99%

8.3 STATISTICAL ANALYSES

Characteristics according to age, gender, and time since diagnosis of COVID-19 will be summarized. Associations will be summarized with chi-square statistics. Logistic regression will be used to study the association of multiple factors with selected conditions at study entry, e.g. pulmonary measurements. Logistic regression will also be used to determine whether the prevalence of medical complications at baseline for COVID-19 survivors differs from controls after adjusting for factors such as age and gender.

Various measures of immune response will be compared between COVID-19 survivors and controls. Depending on the type of measurement, comparisons may be made using the t-test or Fisher's exact test. Multivariate regression will be used for adjusting for potential confounders. Where appropriate, longitudinal data may be analysed using generalized estimating equations.

The rate of medical complications during follow-up will be summarized with life-table methods. Cox regression analysis will be used to study predictors of major outcomes during follow-up among just COVID-19 survivors. Similar analyses will be carried out for comparisons of COVID-19 survivors with controls.

Seroprevalence rates for antibodies to SARS-CoV-2 virus in enrolled controls will be estimated. Trajectories over follow-up of antibody levels of COVID-19 survivors, and of enrolled controls, will be summarized. Predictors of antibody changes will be studied with longitudinal regression models.

Rates of psychiatric diagnoses, including post-traumatic stress disorder (PTSD) as related to COVID-19 diagnosis and treatment experience, will be estimated. Trajectories over follow-up of mental health symptomatology of COVID-19 survivors and controls will be summarized. Predictors of mental health sequelae at follow up will be studied with longitudinal regression models.

To further assess the generalizability of the COVID-19 survivors who enroll in this study, in addition to comparing demographic information of COVID-19 survivors to in-study controls, the survivors may also be compared to demographics from other United States data sets.

In the event that new infections occur during the study, it may be of interest to analyze re-infected and/or newly infected individuals by comparing these individuals to themselves prior to infection, and also by comparing them to other participants in the study using analyses analogous to those described above.

As a descriptive study, no adjustments for multiple comparisons are planned. Results will be reported in a manner that emphasizes point estimates and confidence intervals. If thresholds for statistical significance are required, the Benjamini-Hochberg procedure to control the false discovery rate at 10% will be used.

9 REGULATORY AND OPERATIONAL CONSIDERATIONS

9.1 INFORMED CONSENT PROCESS

9.1.1 Consent/Assent Procedures and Documentation

Informed consent is a process where information is presented to enable persons to voluntarily decide whether or not to participate as a research participant. It is an ongoing conversation between the human research participant and the researchers which begins before consent is given and continues until the end of the participant's involvement in the research. Discussions about the research will provide essential information about the study to include purpose, duration, experimental procedures, alternatives, risks and benefits. Participants will be given the opportunity to ask questions and have them answered. Participants will also have the opportunity to consult others of their choosing (e.g. family members, private physician) prior to providing consent.

The consent process will take place either by telephone (see 9.1.3) or at the NIH CC during the Screening/Baseline visit. Members of the study authorized to obtain informed consent are listed in the Key Study Personnel document. Participants will sign the informed consent document prior to undergoing any research procedures. The participants may withdraw consent at any time throughout the course of the trial. A copy of the informed consent document will be given to the participants for their records. The researcher will document the signing of the consent form in the participant's medical record. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

9.1.2 Consent for minors when they reach the age of majority

N/A

9.1.3 Telephone consent

The informed consent document will be sent to the subject. An explanation of the study will be provided over the telephone after the subject has had the opportunity to read the consent form. The subject will sign and date the informed consent. Signatures may be obtained electronically using a 21 CFR Part 11 compliant platform like DocuSign (e.g. subjects will use a finger or stylus/mouse to create a signature on an electronic informed consent document.)

The original informed consent document will be sent back to the consenting investigator who will sign and date the consent form with the date the consent was returned.

A fully executed copy will be returned via mail or secure email for the subject's records.

The informed consent process will be documented in the medical record.

The investigator will confirm that, when required, written legally effective consent has been obtained prior to initiating any study interventions.

9.1.4 Telephone assent

N/A

9.1.5 Considerations for Consent of NIH employees

Consent for NIH employees will be obtained as detailed above with following additional protections:

Consent from employees will be obtained by an individual independent of the employee's team whenever possible. Otherwise, the consent procedure will be independently monitored by the CC Department of Bioethics Consultation Service in order to minimize the risk of undue pressure on the employee.

9.1.6 Consent of Participants who are/become Decisionally Impaired

Participants who are decisionally impaired and lack the ability to consent will not be enrolled on to this study. Should a participant become decisionally impaired during participation, the following procedures will be followed: The PI or AI will contact the NIH Ability to Consent Assessment Team (ACAT) for evaluation as needed for the following: an independent assessment of whether an individual has the capacity to provide consent; assistance in identifying and assessing an appropriate surrogate when indicated; and/or an assessment of the capacity to appoint a surrogate. For those participants that become incapacitated and do not have pre-determined substitute decision maker, the procedures described in NIH HRPP SOP 14E for appointing a surrogate decision maker for adult participants who are (a) decisionally impaired, and (b) who do not have a legal guardian or durable power of attorney, will be followed.

9.2 STUDY DISCONTINUATION AND CLOSURE

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to study participants. If the study is prematurely terminated or suspended, the PI will promptly inform study participants, the Institutional Review Board (IRB) and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination that the primary objective has been met

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the IRB.

9.3 CONFIDENTIALITY AND PRIVACY

Participant confidentiality and privacy is strictly held in trust by the participating investigators, and their staff. This confidentiality is extended to cover testing of biological samples and to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence.

All research activities will be conducted in as private a setting as possible.

The Institutional Review Board (IRB), and/or regulatory agencies may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be stored at the National Institutes of Health (NIH), Bethesda, MD. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by NIH research staff will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at the NIH.

To further protect the privacy of study participants, a Certificate of Confidentiality has been issued by the NIH. This certificate protects identifiable research information from forced disclosure. It allows the investigator and others who have access to research records to refuse to disclose identifying information on research participation in any civil, criminal, administrative, legislative, or other proceeding, whether at the federal, state, or local level. By protecting researchers and institutions from being compelled to disclose information that would identify research participants, Certificates of Confidentiality help achieve the research objectives and promote participation in studies by helping assure confidentiality and privacy to participants.

9.4 FUTURE USE OF STORED SPECIMENS AND DATA

- **Intended Use:** Stored blood samples and data collected under this protocol may be used to study clinical, immunological, and immunologic parameters of COVID-19. Samples may also be used to study other aspects of the immunopathogenesis of COVID-19.
- **Storage:** Access to stored samples will be limited using a locked room or a locked freezer. Samples and data will be stored using codes assigned by the investigators. Data will be kept in password-protected computers. Only investigators will have access to the samples and data. Audio recordings will be digital and identified by study number without other identifiers. These recordings will be stored on secure servers that are password protected and only members of the research team will have access to them. Recordings will be destroyed with the research is complete.

- **Tracking:** Samples will be tracked utilizing the repository operated by Leidos Biomedical, Inc. Data will be stored and maintained in the NIAID CRIMSON database.
- **Disposition at the Completion of the Protocol:** In the future, other investigators (both at NIH and outside) may wish to use these samples and/or data for research purposes. If the planned research falls within the category of "human subjects research" on the part of the NIH researchers, IRB review and approval will be obtained. This includes the NIH researchers sending out coded and linked samples or data and getting results that they can link back to their subjects.
- **Reporting the Loss or Destruction of Samples/Specimens/Data to the IRB:**
    - Any loss or unanticipated destruction of samples or data (for example, due to freezer malfunction) that meets the definition of Protocol Deviation and/or compromises the scientific integrity of the data collected for the study, will be reported to the IRB.
    - Additionally, participants may decide at any point not to have their samples stored. In this case, the PI will destroy all known remaining samples and report what was done to both the subject and to the IRB. This decision will not affect the subject's participation in other protocols at NIH.

9.5 SAFETY OVERSIGHT

The clinical research team will meet on a regular basis (approximately weekly) when participants are being actively enrolled/evaluated on the study to discuss each participant.

All data will be collected in a timely manner and reviewed by the PI or a lead associate investigator. Events meeting requirements for expedited reporting as described in HRPP Policy 801 will be submitted within the required timelines.

The PI will review all data on each participant to ensure safety and data accuracy. The PI will personally conduct or supervise the investigation and provide appropriate delegation of responsibilities to other members of the research staff.

9.6 CLINICAL MONITORING

This is a cohort natural history study and participants may have very complicated and involved underlying medical problems at baseline. The only research procedures in this study are phlebotomy, leukapheresis, pulmonary function testing, chest x-ray and completion of questionnaires. All Standard diagnostic studies and consultations will be performed as clinically indicated in accordance with standard medical practice for evaluation of symptoms and abnormal physical findings. Therefore, only AEs that occur within 48 hours of phlebotomy, leukapheresis, pulmonary function testing, and completion of questionnaires will be monitored in this protocol.

Information on adverse events possibly or definitely related to of phlebotomy, leukapheresis, pulmonary function testing, and completion of questionnaires will be collected by the study team and entered into the CRIMSON database. This data will be reviewed on an ongoing basis by the PI. On an annual basis, 10% of the Informed Consent Documents and Eligibility Checklists of subjects enrolled in the previous calendar year will be reviewed by the PI and Study Coordinator for completeness and accuracy.

9.7 QUALITY ASSURANCE AND QUALITY CONTROL

The study team will perform internal quality management of study conduct, data and biological specimen collection, documentation and completion.

9.8 DATA HANDLING AND RECORD KEEPING

9.8.1 Data Collection and Management Responsibilities

Data collection is the responsibility of the clinical trial staff under the supervision of PI. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

Study data will be maintained in the NIAID CRIMSON database system and collected directly from subjects during study visits and telephone calls or will be abstracted from subjects' medical records. Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary to confirm the data abstracted for this study. Data entry into CRIMSON will be performed by authorized individuals. Corrections to CRIMSON shall be tracked electronically with time, date, individual making the correction, and what was changed.

The investigator is responsible for assuring that the data collected are complete, accurate, and recorded in a timely manner.

9.8.2 Study Records Retention

Study documents should be retained for a minimum of 2 years after the last approval of a marketing application in an International Conference on Harmonization (ICH) region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the study intervention, or as per the NIH Intramural Records Retention Schedule. No records will be destroyed without the written consent of the sponsor, if applicable. It is the responsibility of the sponsor to inform the investigator when these documents no longer need to be retained.

9.9 UNANTICIPATED PROBLEMS

9.9.1 Definition of Unanticipated Problems (UP)

Any incident, experience, or outcome that meets **all** of the following criteria:

- Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the Institutional Review Board (IRB)-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied; and

- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
- Suggests that the research places participants or others (which many include research staff, family members or other individuals not directly participating in the research) at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or expected.

9.9.2 Unanticipated Problem Reporting

The investigator will report unanticipated problems (UPs) to the NIH Institutional Review Board (IRB) as per Policy 801.

9.10 PROTOCOL DEVIATIONS

It is the responsibility of the investigator to use continuous vigilance to identify and report deviations to the NIH Institutional Review Board as per Policy 801. All deviations must be addressed in study source documents. The investigator is responsible for knowing and adhering to the reviewing IRB requirements.

9.10.1 NIH Definition of Protocol Deviation

A protocol deviation is any changed, divergence, or departure from the IRB-approved research protocol.

- Major deviations: Deviations from the IRB approved protocol that have, or may have the potential to, negatively impact the rights, welfare or safety of the participant, or to substantially negatively impact the scientific integrity or validity of the study.
- Minor deviations: Deviations that do not have the potential to negatively impact the rights, safety or welfare of participants or others, or the scientific integrity or validity of the study.

9.11 PUBLICATION AND DATA SHARING POLICY

9.11.1 Human Data Sharing Plan

This study will be conducted in accordance with the following publication and data sharing policies and regulations:

National Institutes of Health (NIH) Public Access Policy, which ensures that the public has access to the published results of NIH funded research. It requires scientists to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication.

This study will comply with the NIH Data Sharing Policy and Policy on the Dissemination of NIH-Funded Clinical Trial Information and the Clinical Trials Registration and Results Information Submission rule. As such, this trial will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov. In addition, every attempt will be made to publish results in peer-reviewed journals. Data from this study may be requested from other researchers 5 years after the completion of the study.

9.11.2 Genomic Data Sharing Plan

N/A

9.12 COLLABORATIVE AGREEMENTS

9.12.1 Agreement Type

N/A

9.13 CONFLICT OF INTEREST POLICY

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial. The study leadership in conjunction with the NIAID has established policies and procedures for all study group members to disclose all conflicts of interest and will establish a mechanism for the management of all reported dualities of interest.

10 ABBREVIATIONS

AE	Adverse Event
ANCOVA	Analysis of Covariance
CFR	Code of Federal Regulations
CLIA	Clinical Laboratory Improvement Amendments
CMP	Clinical Monitoring Plan
CC	Clinical Center
COVID-19	Coronavirus disease 2019
CRF	Case Report Form
DCC	Data Coordinating Center
DSMB	Data Safety Monitoring Board
EC	Ethics Committee
EUA	Emergency Use Authorization

eCRF	Electronic Case Report Forms
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practices
GMP	Good Manufacturing Practices
GWAS	Genome-Wide Association Studies
ICH	International Conference on Harmonization
ICMJE	International Committee of Medical Journal Editors
IRB	Institutional Review Board
ISM	Independent Safety Monitor
MOP	Manual of Procedures
mSv	millisievert
NIH	National Institutes of Health
NIAID	National Institute of Allergy and Infectious Diseases
OHRP	Office for Human Research Protections
PI	Principal Investigator
QA	Quality Assurance
QC	Quality Control
SAE	Serious Adverse Event
SARS-CoV-2	Severe acute respiratory syndrome coronavirus 2
UP	Unanticipated Problem
US	United States

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