NCT04359810

Protocol (Population A) V 17AUG2020

Long title: A Phase 2, Randomized Clinical Trial to Evaluate the Efficacy and Safety of Human Anti-SARS-CoV-2 Convalescent Plasma in Severely Ill Adults with COVID-19 (AAAS9924)

Short title: Population A: Convalescent Plasma in Severe COVID-19 Clinical

Phase: 2 IND Sponsor: Andrew B. Eisenberger, MD

Principal Investigator: Max O'Donnell, MD, MPH

Conducted by: Columbia University

Sample Size: 220

Study Population: Subjects aged ≥18 years with severe COVID-19

Study Duration: Approximately 1 year.

Study Design: Randomized blinded phase 2 trial to assess the efficacy and safety of anti-SARS-CoV-2 plasma among adults with severe COVID-19. A total of 220 eligible subjects will be randomized in a 2:1 ratio, stratified by country (US, Brazil) to receive either convalescent plasma qualitatively positive for SARS-CoV-2 antibody (anti-SARS-CoV-2 plasma) or non-convalescent fresh frozen plasma (control plasma). The patient and the study clinician assessing the clinical outcome will be blinded to the treatment arm.

The following will be assessed in all subjects:

Safety and efficacy: Day 0 (baseline) to Day 28.

Study Agent:

• Anti-SARS-CoV-2 convalescent plasma (1 unit; ~200-250 mL collected by apheresis from a volunteer who recovered from COVID-19 (collection and qualification covered by IRB protocol AAAS9845 (Convalescent plasma donors)) and was found to be a qualitatively positive for SARS-CoV-2 antibody ("anti-SARS-CoV-2 plasma").

• Control plasma: 1 unit of standard plasma collected prior to December 2019 Protocol (Population A) V 17AUG2020

Primary Efficacy Objective: To evaluate the efficacy of treatment with anti-SARS-CoV-2 plasma versus control plasma with respect to Day 28 severity outcome.

Primary Endpoint: The primary endpoint is Day 28 severity outcome on a seven-category scale:
1. Not hospitalized with resumption of normal activities
2. Not hospitalized, but unable to resume normal activities
3. Hospitalized, not requiring supplemental oxygen
4. Hospitalized, requiring supplemental oxygen
5. Hospitalized, requiring high-flow oxygen therapy or noninvasive mechanical ventilation
6. Hospitalized, requiring extracorporeal membrane oxygenation (ECMO), invasive mechanical ventilation, or both
7. Death

Primary Safety Objective: To evaluate the safety and tolerability of treatment with anti- SARS-CoV-2 plasma versus control (control plasma) in adults with severe COVID-19.

Primary Safety Endpoints:

1. Cumulative incidence of grade 3 and 4 adverse events during the study period
2. Cumulative incidence of serious adverse events during the study period

Secondary Objectives:

1. To compare duration of need for supplemental oxygen and/or mechanical ventilation between recipients of the anti-SARS-CoV-2 plasma and control plasma.
2. To compare duration of hospitalization between recipients of the anti-SARSCoV-2 plasma and control plasma.
3. To compare in-hospital and 28-day mortality between recipients of the anti-SARS-CoV-2 plasma and control plasma.

Ancillary Endpoints:

1. To compare the proportion and duration of SARS-CoV-2 PCR positivity (RT PCR) between the recipients of the anti-SARS-CoV-2 plasma and control plasma at days 0, 7, and 14.
2. To compare the levels of SARS-CoV-2 RNA between recipients of the anti0SARS-CoV-2 plasma and control plasma at days 0, 7, and 14.
3. To assess for genetic and transcriptomic differences at Day 0 (genomic) and Day 0,7,14 (transcriptomic) between the recipients of the anti-SARS-CoV-2 plasma and control plasma.

STUDY POPULATION

1.1.1. Inclusion Criteria for Enrollment

1. Willing and able to provide written informed consent prior to performing study procedures or have a legally authorized representative available to do so.

2. Age ≥18 years.

3. Evidence of SARS-CoV-2 infection by PCR test of nasopharyngeal swab or oropharyngeal swab/tracheal aspirate sample within 14 days of randomization.

4. Peripheral capillary oxygen saturation (SpO2) ≤ 94% on room air or requiring supplemental oxygen, non-invasive or invasive mechanical ventilation at screening.

5. Evidence of infiltrates on chest radiography.

6. Females of childbearing age and males must be willing to practice an effective contraceptive method or remain abstinent during the study period.

1.1.2. Exclusion Criteria

1. Participation in another clinical trial of anti-viral agent(s)* for COVID-19.

2. Receipt of any anti-viral agent(s)* with possible activity against SARS-CoV-2 <24 hours prior to study drug administration.

3. Mechanically ventilated (including veno-venous (VV)-ECMO) ≥ 5 days.

4. Severe multi-organ failure.

5. History of prior reactions to transfusion blood products meeting definitive case definition criteria, at least severe severity, and probable or definite imputability per National Healthcare Safety Network (NHSN)/Centers for Disease Control and Prevention (CDC) criteria.

6. Known IgA deficiency.

7. Females who are pregnant or breastfeeding.

* Use of Remdesivir as treatment for COVID-19 is permitted.

BACKGROUND AND SCIENTIFIC RATIONALE

There are currently no proven treatment options for coronavirus disease (COVID-19), which is caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). Human convalescent plasma has been successfully used for treatment of other severe coronavirus infections and thus may provide an option for treatment of COVID-19 and could be rapidly available from people who have recovered from disease and can donate plasma. Passive antibody therapy involves the administration of antibodies against a given infectious agent to a susceptible or ill individual for the purpose of preventing or treating an infectious disease caused by that agent. Experience from prior outbreaks with other coronaviruses, such as SARS-CoV-1, shows that convalescent plasma contains neutralizing antibodies to the relevant virus (Zhang et al., 2005). In the case of SARS-CoV-2, the anticipated mechanism of action by which passive antibody therapy would mediate protection is viral neutralization. However, other mechanisms may be possible, such as antibody dependent cellular cytotoxicity and/or phagocytosis. Convalescent serum was also used in the 2013 African Ebola epidemic. A small 73 non-randomized study in Sierra Leone revealed a significant increase in survival for those 74 treated with convalescent whole blood relative to those who received standard treatment (Sahr et al., 2017). The only antibody type that is currently available for immediate use against SARS-CoV-2 is that found in human convalescent plasma. As more individuals contract COVID-19 and recover, the number of potential donors will continue to increase. When used for therapy, antibody is most effective when administered shortly after the onset of symptoms. The reason for temporal variation in efficacy is not well understood but could reflect that passive antibody works by neutralizing the initial inoculum, which is likely to be much smaller than that of established disease. Another explanation is that antibody works by modifying the inflammatory response, which is also easier during the initial immune response, which may be asymptomatic (Casadevall & Pirofski, 2003). As an example, passive antibody therapy for pneumococcal pneumonia was most effective when administered shortly after the onset of symptoms and there was no benefit if antibody administration was delayed past the third day of disease (Casadevall & Scharff, 1994). For passive antibody therapy to be effective, a sufficient amount of antibody must be administered. When given to a susceptible person, this antibody will circulate in the blood, reach tissues and provide protection against infection. Depending on the antibody amount and composition, the protection conferred by the transferred immunoglobulin can last from weeks to months.

Experience with the use of convalescent plasma against coronavirus diseases

In the 21st century, there were two other epidemics with coronaviruses that were associated with high mortality, SARS1 in 2003 and MERS in 2012. In both outbreaks, the high mortality and absence of effective therapies led to the use of convalescent plasma. The largest study involved the treatment of 80 patients in Hong Kong with SARS (Cheng et al., 2005). Patients treated before day 14 had improved prognosis defined by discharge from hospital before day 22, consistent with the observation that earlier administration is more likely to be effective. In addition, those who were PCR positive and seronegative for coronavirus at the time of therapy had improved prognosis. There is also some anecdotal information on the use of convalescent plasma in seriously ill individuals. Three patients with SARS in Taiwan were treated with 500 ml of convalescent plasma, resulting in a reduction in plasma virus titer and each survived (Yeh et al., 2005). Three patients with MERS in South Korea were treated with convalescent plasma, but only two of the recipients had neutralizing antibody in their plasma (Ko et al., 2018). The latter study highlights a challenge in using convalescent plasma, namely, that some who recover from viral disease may not have high titers of neutralizing antibody (Arabi et al., 2016).

Consistent with this point, an analysis of 99 samples of convalescent sera from patients with MERS showed that 87 had neutralizing antibody with a geometric mean titer of 1:61. This suggests that antibody declines with time and/or that few patients make high titer responses. It is also possible that other types of non-neutralizing antibodies are made that contribute to protection and recovery as described for other viral diseases (Gunn et al., 2018; van Erp, Luytjes, Ferwerda, & van Kasteren, 2019). There are reports that convalescent plasma was used for therapy of patients with COVID-19 in China during the current outbreak (Xinhua, 2020). Although few details are available from the Chinese experience and published studies involved small numbers of patients, the available information suggests that convalescent plasma administration reduces viral load and was safe.

Overview of known potential risks

Historical and current anecdotal data on use of convalescent plasma suggest it is safe in coronavirus infection. Therefore, the large number of exposed healthcare workers, public servants and first responders, in combination with high morbidity and mortality in severe COVID-19, particularly in elderly and vulnerable persons, suggest that the benefits of convalescent plasma outweigh its possible risks in patients with severe illness. However, for all cases where convalescent plasma administration is considered, a risk-benefit assessment must be conducted to assess individual variables.

The theoretical risk involves the phenomenon of antibody-mediated enhancement of infection (ADE). ADE can occur for several viral diseases and involves an enhancement of disease in the presence of certain antibodies. For coronaviruses, several mechanisms for ADE have been described and there is the theoretical concern that antibodies to one type of coronavirus could enhance infection to another viral strain (Wan et al., 2020). It may be possible to predict the risk of ADE of SARS-CoV-2 experimentally, as proposed for MERS. Since the proposed use of convalescent plasma in the COVID-19 epidemic would rely on preparations with high titers of neutralizing antibody against the same virus, SARS2-CoV-2, ADE may be unlikely. The available evidence from the use of convalescent plasma in patients with SARS1 and MERS (Mair-Jenkins et al., 2015), and anecdotal evidence of its use in patients with COVID-19 (Xinhua, 2020), suggest it is safe. Nevertheless, caution and vigilance will be required in for any evidence of enhanced infection. Another theoretical risk is that antibody administration to those exposed to SARSCoV-2 may avoid disease but modify the immune response such that those individuals mount attenuated immune responses, which would leave them vulnerable to subsequent re-infection. In this regard, passive antibody administration before vaccination with respiratory syncytial virus was reported to attenuate humoral but not cellular immunity (Crowe, Firestone, & Murphy, 2001). This concern will be investigated as part of this clinical trial by measuring immune responses in those exposed and treated with convalescent plasma to prevent disease. If the concern proved real these individuals could be vaccinated against COVID-19 when a vaccine becomes available. Passive antibodies are derived from human serum. The antibodies used in this study will be derived from serum obtained from convalescent patients, and will be subjected to testing protocols that are similar to those used by blood banks and transfusion services. However, as is the case with any biological product, there is a very small risk of allergy/anaphylaxis, or passive transfer of potential unknown infectious agents or infections. While most adverse effects are mild and transient including headaches, flushing, fever, chills, fatigue, nausea, diarrhea, transient changes in blood pressure and tachycardia, there is also the risk of transfusion related acute lung injury (TRALI), and transfusion associated circulatory overload (TACO), which could worsen hypoxemia in patients requiring supplemental oxygen or non-invasive or mechanical ventilation. Late adverse events are rare and include acute renal failure and thromboembolic events.

Known potential benefits

A benefit of convalescent plasma administration is that it can prevent infection and subsequent disease in those who are at high risk for disease following close contacts of patients with COVID-19. This is especially so for those with underlying medical conditions. Many who will qualify for prophylaxis are health care workers and first responders who are critical to maintenance of stability of the healthcare system.

Passive antibody administration to prevent disease is already used in clinical practice. For example, patients exposed to hepatitis B and rabies viruses are treated with hepatitis B immune globulin (HBIG) and human rabies immune globulin (RIG), respectively. Botulism Immune Globulin Intravenous (Human) (BIG-IV) is an intravenous preparation for infant botulism. In addition, passive antibody is used for the prevention of severe respiratory syncytial virus (RSV) disease in high-risk infants. Until recently, polyclonal hyperimmune globulin (RSV-IG) prepared from donors selected for having high plasma titers of RSV neutralizing antibody, was used but these preparations have now been replaced by palivizumab, a humanized murine monoclonal antibody. Another potential benefit is societal: If the frequency with which exposed persons become infected decreases, the risk of further transmission (R naught) might be reduced and the epidemic slowed. Another avenue (not pursued in this protocol) is as a treatment for established infection. Convalescent plasma would be administered to those with clinical disease in an effort to reduce their symptoms and mortality. Based on the historical experience with antibody administration, it can be anticipated that antibody administration would be more effective in preventing disease than in the treatment of established disease. However, potential benefits in patients with known infection include reduced severity of symptoms, reduced duration of hospitalization, reduced likelihood of death due to infection, and increased speed of recovery. Given that historical and current anecdotal data on use of convalescent plasma suggest it is safe in coronavirus infection, the high mortality of COVID-19, particularly in elderly and vulnerable persons, suggests that the benefits of its use in those at high risk for or with early disease outweigh the risks. However, for all cases where convalescent plasma administration is considered, a risk-benefit assessment must be conducted to assess individual variables.

INVESTIGATIONAL PLAN

Study Objectives

Primary Objectives:

Primary Efficacy Objective: To evaluate the efficacy of treatment with anti-SARS-CoV-2 plasma versus control plasma with respect to Day 28 severity outcome on a seven-category ordinal scale.
Primary Safety Objective: To evaluate the safety and tolerability of treatment with anti- SARS-CoV-2 plasma versus control plasma in adults with severe COVID-19.

Secondary Objectives:

1. To compare duration of need for supplemental oxygen and/or mechanical ventilation between recipients of the anti-SARS-CoV-2 plasma and control plasma.
2. To compare duration of hospitalization between recipients of the anti-SARS-CoV-2 plasma and control plasma.

3. To compare in-hospital and 28-day mortality between recipients of the anti-SARS-CoV-2 plasma and control plasma.

Ancillary Endpoints:

1. To compare the proportion and duration of SARS-CoV-2 PCR positivity (RT PCR) between recipients of the anti-SARS-CoV-2 plasma and control plasma at days 0, 7, and 14.
2. To compare the levels of SARS-CoV-2 RNA between recipients of the anti-SARS-CoV-2 plasma and control plasma at days 0, 7, and 14.
3. To compare genetic differences between recipients of the anti-SARS-CoV-2 plasma and control plasma to explore possible associations with poor outcome.
4. To compare transcriptomic differences between recipients of the anti-SARS-CoV-2 plasma and control plasma at Day 0, 7, and 14 to explore possible associations with poor outcome.

Definitions

Enrolled: From time consented to participate until designated as a screen failure or have either been discontinued from the study or completed it. II. Randomized: when a study arm is assigned. III. Screen Failures: signed informed consent, but then determined to be ineligible or withdraws before being randomized. IV. Discontinued: randomized, but then withdrawn by investigator or withdraws consent. V. Completed: Subjects are considered completed when they are followed through to day 28, if they die before day 28, or are discharged prior to day 14.

Study population

Inclusion Criteria for Enrollment 1. Willing and able to provide written informed consent prior to performing study procedures or having a legally authorized representative available to do so. 2. Age ≥18 years. 3. Evidence of SARS-CoV-2 infection by PCR test of nasopharyngeal swab or oropharyngeal swab/tracheal aspirate sample within 14 days of randomization 4. SPO2 ≤ 94% on room air or requiring supplemental oxygen, non-invasive or invasive mechanical ventilation at screening 5. Evidence of infiltrates on chest radiography 6. Females of childbearing age and males must be willing to practice an effective contraceptive method or remain abstinent during the study period.

Exclusion Criteria for Enrollment 1. Participation in another clinical trial of anti-viral agent(s)* for COVID-19 2. Receipt of any anti-viral agent(s)* with possible activity against SARS-CoV-2 < 24 hours prior to plasma infusion. 3. Mechanically ventilated (including VV-ECMO) ≥ 5 days 4. Severe multi-organ failure 5. History of allergic reactions to transfusion blood products per NHSN/CDC criteria 6. Known IgA deficiency 7. Females who are pregnant or breastfeeding. *Use of remdesivir as treatment for COVID-19 is permitted.

Subject Withdrawal

Subjects can terminate study participation and/or withdraw consent at any time without prejudice. II. Randomized subjects who withdraw from the study will not be replaced. III. The investigator may withdraw subjects if they are lost to follow up, non-compliant with study procedures or if the investigator determines that continued participation in the study would be harmful to the subject or the integrity of the study data. IV. Discontinuation of the study: The study sponsor, FDA and IRB all have the right to terminate this study at any time.

Intervention

Subjects will be randomized in a 2:1 ratio, stratified by country, to receive treatment vs. frozen fresh plasma. II. Study drug: The investigational product is anti-SARS-CoV-2 plasma. Patients identified as having recovered from COVID-19 will serve as potential donors. Testing will confirm presence of anti-SARS-CoV-2 antibody prior to donation. Potential donors and samples will be screened for transfusion-transmitted infections (e.g. HIV, HBV, HCV, WNV, HTLV-I/II, T. cruzi, ZIKV) and plasma will be collected using apheresis technology. This is similar to standard blood bank protocols. III. Active arm will receive 1 unit of anti-SARS-CoV-2 plasma. IV. Control arm will receive 1 unit of control plasma. V. The study drug will be in a standard plasma unit bag, with a studyspecific ISBT label and will include the following statement: “Caution: New Drug--Limited by Federal (or United States) law to investigational use." VI. The blood bank will not be blinded to treatment allocation, nor will the clinical study team. VII. The patient and the clinician who will assess the end of treatment outcome will be blinded to treatment allocation.

Randomization

Subjects enrolled in the study will be randomized to receive study drug vs. control using a web-based randomization platform that will pre-generate all treatment assignments in a 2:1 ratio using random permuted blocks of random block sizes, stratified by country (US and Brazil) The assignment list is maintained by designated staff at the DCC, independent from the study, and then sent to the Principal Investigator/research team for each participant that is deemed eligible.

Study product considerations

The preparation of the anti-SARS plasma and the control plasma will take place at the New York City Blood Center and the CUIMC-NYPH Blood Bank will dispense the plasma products. The plasma collection procedures are not part of this research protocol and are described in separate protocols, which has separate IRB approval (AAAS9845). The description below provides a summary of study product considerations as context.

Collection:

All activities pertaining to the collection and processing of plasma will take place at [New York Blood Center/NYBC]. NYBC is one of the largest independent, community-based, nonprofit blood centers in the United States. NYBC has a longstanding research program and is well versed in the regulatory and ethical aspects of research, including clinical trials. The organization is FDA-licensed to produce convalescent plasma and AABB (American Association of Blood Banks) accredited, attesting to robust quality oversight of all operations. The donation and collection of donor convalescent plasma will occur under CUIMC IRB protocol AAAS9845.

Collection and processing:

• Standard apheresis plasma collection will be performed per routine standard operating procedure at the collection facility (NYBC). • As per routine practice, samples will be collected at time of donation for testing for transfusion-transmissible infections (all donors), ABO and red cell antibodies (all donors) and HLA antibodies (female donors with prior pregnancies). • Target collection volume: ~450-600mL; this will allow for later splitting (separation) into 200-250mL daughter units. • The plasma will be processed

per routine practice; it will be frozen within 24hrs of collection per AABB standards. • The plasma will be maintained in quarantine at the blood center pending laboratory test results (i.e. infectious screening, ABO and RhD status, Red cell and HLA antibodies). • If laboratory testing is acceptable (i.e. negative infectious and antibody screening), the products will be distributed to hospital blood bank for storage. • In the event of an abnormal test result, the product will be discarded and the donor will be notified by the blood center as is standard practice.

Control arm plasma:

The control arm plasma follows identical collection and processing procedures, but will have been collected from community blood donors prior to documented SARS-CoV-2 in the United States (i.e., to be conservative, all control arm plasma will be the oldest available plasma and should be from collections prior to 31 December 2019).

Rationale for dosing:

We will utilize 1 unit (200-250 mL) of plasma with anti-SARS-CoV-2 antibody. Dosing was based on experience with previous use of convalescent plasma therapy in SARS1 where 5 mL/kg of plasma at titer ≥ 1:160 was utilized [European journal of clinical microbiology & infectious diseases: official publication of the European Society of Clinical Microbiology. 2005; 24(1):44- 6.]. Historical precedence allowing for 0.25 of treatment dose was taken into account. Hence, considering first order linear proportionality, 3.125mL/kg of plasma with titer >1:64 would provide equivalent immunoglobulin level to one quarter of 5ml/kg plasma with titer ≥ 1:160. For a typical patient (~80 Kg) this would result in 250 mL of plasma (3.125ml/kg x 80kg = 250 mL > 1:64).

Study drug administration:

• Drug will be administered within 48 hours of randomization. • Infusion rate ≤ 250 mL/hour at physician discretion • Pretreatment to minimize transfusion reactions (e.g. acetaminophen, diphenhydramine) will not be given, but will be available as needed to treat fever or allergic reaction. For severe allergic reactions corticosteroids (e.g., 125mg solu-medrol IV) may be used. For rare severe anaphylaxis, epinephrine will be available. • If an AE develops during infusion, the infusion may be slowed or stopped as per investigator's decision. • Most reactions to plasma are relatively minor and the infusion can generally be continued. Infusion site burning and non-allergic systemic effects can generally be managed with slowing of the infusion. Infusion is generally stopped in cases of itching; participant is treated and then infusion cautiously re-started. • Severe allergic reactions generally require discontinuation of the infusion. These include: • Respiratory compromise: dyspnea, wheezing, stridor, hypoxemia A decrease in systolic blood pressure to < 90 mmHg or >30% decrease from baseline or a diastolic drop of >30% from baseline. • Tachycardia with an increase in resting heart rate to > 130bpm; or bradycardia.

Concomitant medications will be documented on the CRF:

• Prescription medications • Over the counter medications • Herbal treatments/nutritional supplements • Blood products

Prohibited Medications:

Any approved or investigational drug* with established or potential activity against SARS-CoV-2 given within 24 hours of plasma infusion. *Concurrent use of Remdesivir as treatment for COVID-19 during the course of the study is permitted.

STATISTICAL CONSIDERATIONS AND DATA MANAGEMENT

Design Overview

This randomized blinded phase 2 trial will assess the efficacy and safety of anti-SARS-CoV-2 plasma among adults with severe COVID-19. Eligible participants will be randomized in a 2:1 ratio to receive anti-SARS-CoV-2 plasma or fresh frozen plasma without known anti-SARS-CoV-2 antibodies. We plan to enroll up to 220 participants (US and Brazil). Each participant will be evaluated at baseline and daily during the follow-up period using a seven-category severity scale: 1. Not hospitalized with resumption of normal activities, 2. Not hospitalized, but unable to resume normal activities, 3. Hospitalized, not requiring supplemental oxygen, 4. Hospitalized, requiring supplemental oxygen, 5. Hospitalized, requiring high---flow oxygen therapy or noninvasive mechanical ventilation, 6. Hospitalized, requiring extracorporeal membrane oxygenation (ECMO), invasive mechanical ventilation, or both, and 7. Death.

According to the inclusion/exclusion criteria, all enrolled participants will have a score ranging 3—6 on this scale at baseline. The primary study endpoint is defined as the Day 28 outcome on the scale.

Safety will also be evaluated daily. Secondary endpoints include:
• SARS-CoV-2 PCR positivity from nasopharyngeal swab, collected on baseline (Day 0), Days 7, and 14. • Levels of SARS-CoV-2 RNA on Days 0, 7, and 14. • During of need for supplemental oxygen and/or mechanical ventilation • Duration of hospitalization • Mortality.

Statistical Analysis

• Efficacy Objective

1. Primary Analysis, go/no-go decision, and power consideration

The primary analysis of the endpoint will be a one-sided nonparametric stratified Mann-Whitney test; stratification is done by country (US, Brazil). A “go decision” in this phase 2 trial will be a one-sided P<.015, suggesting evidence of promise of the treatment arm for further investigation in a phase 3 trial. Adaptive seamless phase 2/3: The results in this proof-of-concept study will be used to plan the sample size in a Phase 3 trial, which may include the data in this Phase 2 in the final analysis using adjusted P value as in a seamless phase 2/3 trial. Details of P value adjustment and sample size calculation will be decided before the final analysis of this study data.

The calculation was based on blinded pooled data of the Day 28 outcome and an odds ratio of 1.7 under a proportional odds assumption. With a 2:1 randomization ratio and a total sample size of 219 participants (146 in convalescent plasma vs. 73 control), a onesided Mann Whitney test at 15% level will have about 82% power to detect an odds ratio of 1.7. Since the difference in power between 219 and 220 is minimal, N=219 can be used to justify an enrollment total of 220. We note a recent study of antiviral drug yields an odds ratio of 1.15, with 95% CI: 0.67 to 1.96, which covers our assumed odds ratio.

2. Secondary and Ancillary Analyses

All secondary and ancillary efficacy analyses will also be intent-totreat. With a sample size of 220, assuming 20% mortality up top day 28, we expect a total of 43 events. Longitudinal data collected over multiple days during the study period (e.g., PCR positivity, RNA) will be analyzed using the framework of generalized linear mixed model. Time-to-event variables (e.g., time to death) will be analyzed using Cox proportional hazards model. Continuous variables (e.g. duration of hospitalization) will be analyzed using Mann Whitney test. Treatment effects on these variables will be estimated with 95% confidence intervals.

3. Safety Analyses

Serious adverse events will be summarized by grades and types using proportions and 95% confidence intervals for the two study arms. Relative safety profile of the two arms will be compared using Fisher's exact test.

4. Missing data and non-compliance

We will compare the missing data patterns between the study arms; and perform sensitivity analyses using different imputation approaches. However, due to the short study period, we anticipate minimal missing data. All efficacy analyses will be done intent-to-treat, although as-treat analyses will also be conducted as sensitivity.

STUDY PROCEDURES

Screening

A. Screening (must be completed before randomization)
B. Informed consent (obtained before performing study related activities)
C. Baseline Evaluation (at screening) 1. Demographics (age, sex, ethnicity, race) 2. Medical history (acute and chronic medical conditions, medications, allergies) (any medical condition arising after consent should be recorded as AE) 3. COVID-19 symptom screen (fever, cough, shortness of breath) 4. Confirmation of SARS-CoV-2 testing (RT-PCR) for eligibility (within 14 days of randomization) 5. Vital signs 6. Physical examination 7. Blood typing 8. Urine or serum pregnancy test for females of childbearing potential. Results from laboratory tests obtained up to 7 days before enrollment may be used for the pregnancy test. 9. Determination of eligibility as per inclusion/exclusion criteria

Baseline (Randomization)

1. Randomization of eligible subject 2. Vital signs 3. COVID-19 symptom screen (fevers, cough, shortness of breath) 4. New medical conditions, concomitant medication 5. Assessment of clinical status (using 7-point ordinal outcome scale) 6. CBC, comprehensive metabolic panel (abstracted from routine clinical lab results in electronic medical record) 7. Stored samples for future studies (only if feasible) 8. SARS-CoV-2 testing (RT-PCR) from nasopharyngeal swab (only if feasible)

Day 0 Infusion (Within 48 hours from randomization) 1. Study Plasma Administration: A single unit of plasma will be transfused. Time at start and end of infusion will be recorded and vital signs will be measured immediately prior to infusion, 10-20 minutes after start of infusion, and at completion of

infusion. 2. Vital signs 3. Physical examination (acceptable to use clinician notes from electronic medical record) 4. COVID-19 symptom screen (fevers, cough, shortness of breath) 5. New medical conditions, concomitant medication, AE evaluation

Day 1 ± 1 1. Vital signs 2. COVID-19 symptom screen (fevers, cough, shortness of breath) 3. Assessment of clinical status (using 7-point ordinal outcome scale) 4. New medical conditions, AE evaluation 5. Physical examination (acceptable to use clinician notes from electronic medical record) 6. CBC, comprehensive metabolic panel (abstracted from routine clinical lab results in electronic medical record) 7. Stored samples for future studies (only if feasible)

Day 3 ± 2 1. Vital signs 2. COVID-19 symptom screen (fevers, cough, shortness of breath) 3. Assessment of clinical status (using 7-point ordinal outcome scale) 4. New medical conditions, AE evaluation 5. Physical examination (acceptable to use clinician notes from electronic medical record)

Day 7 ± 2 1. Vital signs 2. COVID-19 symptom screen (fevers, cough, shortness of breath) 3. Assessment of clinical status (using 7-point ordinal outcome scale) 4. New medical conditions, AE evaluation 5. Physical examination (acceptable to use clinician notes from electronic medical record) 6. SARS-CoV-2 testing (RT-PCR) from nasopharyngeal swab (only if feasible) 7. CBC, comprehensive metabolic panel (abstracted from routine clinical lab results in electronic medical record) 8. Stored samples for future studies (only if feasible)

Day 14 ± 2 1. Vital signs 2. COVID-19 symptom screen (fevers, cough, shortness of breath) 3. Assessment of clinical status (using 7-point ordinal outcome scale) 4. New medical conditions, AE evaluation 5. Physical examination (acceptable to use clinician notes from electronic medical record) 6. SARS-CoV-2 testing (RT-PCR) from nasopharyngeal swab (only if feasible) 7. CBC, comprehensive metabolic panel (abstracted from routine clinical lab results in electronic medical record) 8. Stored samples for future studies (only if feasible)

Day 28 ± 3 1. Vital signs 2. COVID-19 symptom screen (fevers, cough, shortness of breath) 3. Assessment of clinical status (using 7-point ordinal outcome scale) 4. New medical conditions, AE evaluation 5. Physical examination (acceptable to use clinician notes from electronic medical record)

EFFICACY, VIROLOGIC AND PK MEASURES

Primary Endpoint:

Day 28 severity outcome on a seven-category ordinal scale consisting of the following categories: 1. Not hospitalized with resumption of normal activities 2. Not hospitalized, but unable to resume normal activities 3. Hospitalized, not requiring supplemental oxygen 4. Hospitalized, requiring supplemental oxygen 5. Hospitalized, requiring high-flow oxygen therapy or noninvasive mechanical ventilation 6. Hospitalized, requiring extracorporeal membrane oxygenation (ECMO), invasive mechanical ventilation, or both 7. Death

Virologic measures:

1. Rates and duration of SARS-CoV-2 PCR positivity (RT PCR) at days 0, 7 and 14.
2. Peak quantity levels of SARS-CoV-2 RNA at days 0, 7 and 14.

RISKS AND BENEFITS

Potential benefits of treatment

The potential benefits of antiviral treatment with anti-SARS CoV-2 plasma in patients with severe COVID-19 is unknown. However, based on available evidence from use of convalescent plasma in SARS-CoV-1 patients, shortened duration of illness and improved mortality are potential benefits.

Potential benefits of clinical monitoring and virologic testing

Subjects enrolled in the study will undergo close virological monitoring that may facilitate improved understanding of viral shedding that may have benefit to the individual, their family and the community at large.

Potential risks of study procedures

1. Risks of plasma: Fever, chills, rash, headache, serious allergic reactions, transmission of infectious agents
2. Transfusion related acute lung injury (TRALI) and transfusion related circulatory overload (TACO), both of which may worsen oxygen saturation and increase work-of-breathing
3. Risks of phlebotomy: local discomfort, bruising, hematoma, bleeding, fainting
4. Total blood draws will not exceed 500 mL
5. Risks of nasopharyngeal swab: local discomfort, vomiting
6. Risks of IV placement: bleeding, infection, thrombosis

Potential risks of genetic testing

Samples obtained for future research may include a search for genetic correlates of COVID-19 susceptibility or severity. Specimens will be labeled by study IDs, rather than names. This information will not be released to participants and will not become part of their medical records. Risks related to discrimination or other problems are deemed highly unlikely.

Alternatives

The alternative to participation in this study is continued standard-of-care clinical management.

Safety monitoring

1. Safety Evaluations: Will assess for the safety of anti-SARS-CoV-2 plasma in terms of treatment emergent adverse events.
2. Clinical evaluations: Vital signs and symptom screen on days 0-28 (or until hospital discharge, whichever is sooner).
3. Laboratory evaluations:
4. Safety laboratory tests (ABO typing, pregnancy testing, CBC and comprehensive metabolic panel) will be performed at the local CLIA-certified hospital clinical laboratory on days 0-14 (or until hospital discharge, whichever is sooner).

Adverse event reporting

An Adverse Event (AE) is any untoward or unfavorable medical occurrence in a human subject administered an investigational product, including any abnormal sign, symptom or disease, temporally associated with the subject's participation in research, whether or not considered related to the subject's participation in the research.

An AE does not include the following: • Medical or surgical procedures such as surgery, endoscopy, tooth extraction, and transfusion. The condition that led to the procedure may be an AE and must be reported. • Preexisting diseases, conditions, or laboratory abnormalities present or detected before the screening visit that do not worsen • Situations where an untoward medical occurrence has not occurred (e.g., hospitalization for elective surgery, social and/or convenience admissions) • Any medical condition or clinically significant laboratory abnormality with an onset date before the investigational product is administered to the subject and not related to a protocol-associated procedure is not an AE. It is considered to be preexisting and should be documented as medical history.

Preexisting events or conditions that increase in severity or change in nature after the subject receives the investigational product will be considered AEs.

Serious Adverse Event (SAE): any adverse event temporarily associated with the subject's participation in research that meets any of the following criteria: • Results in death; • Is life-threatening (places the subject at immediate risk of death from the event as it occurred); • Requires inpatient hospitalization or prolongation of existing hospitalization; • Results in a persistent or significant disability/incapacity; • Results in a congenital anomaly/birth defect; or • Any other adverse event that, based upon appropriate medical judgment, may jeopardize the subjects' health and may require medical or surgical intervention to prevent one of the other outcomes listed in this definition.

Prolonged Hospitalization or Surgery Protocol – any AE that results in prolonged hospitalization should be documented and reported as a SAE. Any condition responsible for surgery should be documented as an AE if the condition meets the criteria for an AE. Neither the condition, prolonged hospitalization nor surgery are reported as an AE in the following circumstances: • Prolonged hospitalization for diagnostic or elective surgical procedures for a preexisting co144ndition. Surgery should not be reported as an outcome of an AE if the purpose of the surgery was elective or diagnostic and the outcome was uneventful. • Prolonged hospitalization for required to allow efficacy measurement for the study.

An Unanticipated Problem (UP) is any incident, experience or outcome involving risk to subjects or others in any human subjects research 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 IRB-approval protocol and informed consent document, and (b) the characteristics of the subject population being studied; • Related or possibly related to participation in such research (i.e., there is a reasonable possibility that the incident, experience or outcome may have been caused by the procedures involved in such research); and • Suggests that the research places subjects or others at a greater risk of harm (including physical, psychological, economic or social harm) than was previously known or recognized

A Suspected Adverse Reaction (SAR) is any AE for which there is a reasonable possibility that it was caused by the drug. Reasonable possibility means that there is evidence to suggest a causal relationship between the drug and the AE. Examples of reasonable possibility are: • A single occurrence of an event that is uncommon and known to be strongly associated with drug exposure. • One or more occurrences of an event that is not commonly associated with drug exposure, but is otherwise uncommon in the

population exposed to the drug. • An aggregate analysis of specific events observed in a clinical trial that indicates that those events occur more frequently in the drug treatment group than in a concurrent or historical control group.

Investigator Reporting Requirements

The Principal Investigator will report all AEs and SAEs to the IND sponsor within 48 hours of becoming aware of the event. The report to the IND sponsor will include the study investigator's preliminary assessment of seriousness, severity and relatedness to the investigational product.

To IRB: Unanticipated Problems (UPs) must be reported promptly, but not later than 7 calendar days following the occurrence of the UP or the Principal Investigator's acquiring knowledge of the UP.

To DSMB: Serious adverse events not constituting an unanticipated problem is to be reported to the DSMB and the IND sponsor. Reporting should occur within 48 hours of knowledge of the SAE occurrence.

IND Sponsor Reporting Requirements

The IND sponsor will report the following SARs to the FDA: • To the FDA, as soon as possible, but no later than 7 calendar days after the SI's initial receipt of the information, any unexpected fatal or life-threatening SAR. • To the FDA and all participating investigators, as soon as possible but no later than 15 calendar days after the S-I determines that information qualifies for reporting, in an IND safety report, any SAR that is both serious and unexpected. • To the FDA and all participating investigators, as soon as possible but no later than 15 calendar days after the S-I determines that the information qualifies for reporting, any findings from epidemiological studies, pooled analysis of multiple studies or clinical studies, whether or not conducted under an IND or by the S-I, that suggest a significant risk in humans exposed to the drug. • To the FDA and all participating investigators, as soon as possible, but no later than 15 calendar days after the S-I determines that the information qualifies for reporting, any findings from animal or in vitro testing, whether or not conducted by the S-I, that suggest a significant risk in humans exposed to the drug. • To the FDA and all participating investigators, as soon as possible, but no later than 15 calendar days after the S-I determines that the information qualifies for reporting, any clinically important increase in the rate of a Serious SAR over that listed in the protocol or Investigator Brochure. • Expected SAEs and AEs should be included in the IND Annual Reports.

Follow-up information to a safety report will be submitted as soon as the relevant information is available. However, if the results of a sponsor's investigation show that an adverse drug experience not initially determined to be reportable are so reportable, the sponsor must report such experience as soon as possible, but no later than 15 calendar days after the determination is made.

SAFETY OVERSIGHT

Monitoring Plan

1. All AEs and SAEs will be reviewed by the study team in real time.
2. A data safety monitoring board (DSMB), composed of independent experts without conflict of interests will be established. The Board will review the study before initiation and quarterly thereafter.

The Board will review study data to evaluate the safety, efficacy, study progress, and conduct of the study.

Study monitoring

As per ICH-GCP 5.18 and FDA 21 CFR 312.50, clinical protocols are required to be adequately monitored by the study sponsor. Monitors will verify that
(1) There is documentation of the informed consent process and signed informed consent documents for each subject
(2) There is compliance with recording requirements for data points
(3) All SAEs are reported as required
(4) Individual subjects' study records and source documents align
(5) Investigators are in compliance with the protocol
(6) Regulatory requirements as per Office for Human Research Protections (OHRP), FDA, and applicable guidelines (ICH-GCP) are being followed.

Halting Criteria for the Study

The study enrollment and dosing will be stopped and an ad hoc review will be performed if any of the specific following events occur or, if in the judgment of the study physician, subject safety is at risk of being compromised:
1. Unexpected death of a dosed subject in relation to infusion
2. Occurrence of a life-threatening allergic/hypersensitivity reaction (anaphylaxis), manifested by bronchospasm with or without urticaria or angioedema requiring hemodynamic support with pressor medications or mechanical ventilation.
3. One subject with an unexpected SAE associated with study product.
4. Two subjects with a Grade 3 or higher toxicity for the same parameter associated with study product.
5. An overall pattern of symptomatic, clinical, or laboratory events that the medical monitor, ISM, or SMC consider associated with study product and that may appear minor in terms of individual events but that collectively may represent a serious potential concern for safety.
6. Any other event(s) which is considered to be a serious adverse event in the good clinical judgment of the responsible physician. This will be appropriately documented.

Furthermore, given that ADE may be an issue with convalescent antibody treatment, out of an abundance of caution we will monitor the number of subjects in each trial arm that progresses to an indication for need of mechanical ventilation. In monitoring the number of subjects that progresses to this stage, we will present these data to the DSMB and a masked outcomes accessor so that they may objectively evaluate and determine whether they would like to be unmasked. After at least 50% of trial participants have accumulated follow-up, the number of subjects that progress to this stage will be presented to the masked outcomes assessor and formally asked whether they (1) see a clinically meaningful difference between trial arms and (2) if so do they require a formal interim analysis. At any point should the DSMB require a formal interim analysis, we will examine the difference in treatment arms for need for mechanical ventilation. This interim analysis will adjust for factors related to need for mechanical ventilation including age and presence of cardiopulmonary comorbidities.

Upon completion of this review and receipt of the advice of the DSMB, the IND sponsor will determine if study entry or study dosing should be interrupted or if study entry and study dosing may continue according to the protocol. Should the trial not be stopped at this time point, the final analysis would

need to account the number of interim analyses that were conducted. Therefore, we would penalize any final analysis dividing our 0.05 alpha in half for each interim analysis.

REFERENCES

- Arabi, Y. M., Hajeer, A. H., Luke, T., Raviprakash, K., Balkhy, H., Johani, S., . . . Alahmadi, M. (2016). Feasibility of Using Convalescent Plasma Immunotherapyfor MERS-CoV Infection, Saudi Arabia. Emerg Infect Dis, 22(9), 1554-1561. doi:10.3201/eid2209.151164
- Austin, P. C., & Fine, J. P. (2017). Practical recommendations for reporting Fine-Gray model analyses for competing risk data. Stat Med, 36(27), 4391-4400. doi:10.1002/sim.7501
- Casadevall, A., & Pirofski, L. A. (2003). Antibody-mediated regulation of cellular immunity and the inflammatory response. Trends Immunol, 24(9), 474-478. doi:10.1016/s1471-4906(03)00228-x
- Casadevall, A., & Scharff, M. D. (1994). Serum therapy revisited: animal models of infection and development of passive antibody therapy. Antimicrob Agents Chemother, 38(8), 1695-1702. doi:10.1128/aac.38.8.1695
- Cheng, Y., Wong, R., Soo, Y. O., Wong, W. S., Lee, C. K., Ng, M. H., . . . Cheng, G. (2005). Use of convalescent plasma therapy in SARS patients in Hong Kong. Eur J Clin Microbiol Infect Dis, 24(1), 44-46. doi:10.1007/s10096-004-1271-9
- Crowe, J. E., Jr., Firestone, C. Y., & Murphy, B. R. (2001). Passively acquired antibodies suppress humoral but not cell-mediated immunity in mice immunized with live attenuated respiratory syncytial virus vaccines. J Immunol, 167(7), 3910-3918. doi:10.4049/jimmunol.167.7.3910
- Fine, J. P., Gray, R. J. (1999). A Proportional Hazards Model for the Subdistribution of a Competing Risk. Journal of the Americal Statistical Association, 94(446), 496- 509. doi:10.1080/01621459.1999.10474144
- Gunn, B. M., Yu, W. H., Karim, M. M., Brannan, J. M., Herbert, A. S., Wec, A. Z., . . . Alter, G. (2018). A Role for Fc Function in Therapeutic Monoclonal AntibodyMediated Protection against Ebola Virus. Cell Host Microbe, 24(2), 221-233 e225. doi:10.1016/j.chom.2018.07.009
- Kahan, B. C., Jairath, V., Dore, C. J., & Morris, T. P. (2014). The risks and rewards of covariate adjustment in randomized trials: an assessment of 12 outcomes from 8 studies. Trials, 15, 139. doi:10.1186/1745-6215-15-139
- Ko, J. H., Seok, H., Cho, S. Y., Ha, Y. E., Baek, J. Y., Kim, S. H., . . . Peck, K. R. (2018). Challenges of convalescent plasma infusion therapy in Middle East respiratory coronavirus infection: a single centre experience. Antivir Ther, 23(7), 617-622. doi:10.3851/IMP3243
- Liang, K., Zeger, S. L. (1986). Longitudinal data analysis using generalized linear models. Biometrika, 73(1), 13-22.
- Mair-Jenkins, J., Saavedra-Campos, M., Baillie, J. K., Cleary, P., Khaw, F. M., Lim, W. S., . . . Convalescent Plasma Study, G. (2015). The effectiveness of convalescent plasma and hyperimmune immunoglobulin for the treatment of severe acute respiratory infections of viral etiology: a systematic review and exploratory meta-analysis. J Infect Dis, 211(1), 80-90. doi:10.1093/infdis/jiu396
- Sahr, F., Ansumana, R., Massaquoi, T. A., Idriss, B. R., Sesay, F. R., Lamin, J. M., . . . Gevao, S. M. (2017). Evaluation of convalescent whole blood for treating Ebola Virus Disease in Freetown, Sierra Leone. J Infect, 74(3), 302-309. doi:10.1016/j.jinf.2016.11.009
- van Erp, E. A., Luytjes, W., Ferwerda, G., & van Kasteren, P. B. (2019). Fc-Mediated Antibody Effector Functions During Respiratory Syncytial Virus Infection and Disease. Front Immunol, 10, 548. doi:10.3389/fimmu.2019.00548

- Wan, Y., Shang, J., Sun, S., Tai, W., Chen, J., Geng, Q., . . . Li, F. (2020). Molecular Mechanism for Antibody-Dependent Enhancement of Coronavirus Entry. J Virol, 94(5). doi:10.1128/JVI.02015-19
- Xinhua. (2020). China puts 245 COVID-19 patients on convalescent plasma therapy. Retrieved from http://www.xinhuanet.com/english/2020-02/28/c_138828177.htm
- Yeh, K. M., Chiueh, T. S., Siu, L. K., Lin, J. C., Chan, P. K., Peng, M. Y., . . . Chang, F. Y. (2005). Experience of using convalescent plasma for severe acute respiratory syndrome among healthcare workers in a Taiwan hospital. J Antimicrob Chemother, 56(5), 919-922. doi:10.1093/jac/dki346 Zhang, J. S., Chen, J. T., Liu, Y. X.,
- Zhang, Z. S., Gao, H., Liu, Y., . . . Yin, W. D. (2005). A serological survey on neutralizing antibody titer of SARS convalescent sera. J Med Virol, 77(2), 147-150. doi:10.1002/jmv.20431