Protocol Title:

Efficacy of Novel Agents for Treatment of SARS-CoV- 2 Infection Among High-Risk Outpatient Adults: An Adaptive Randomized Platform Trial

NCT Number:

NCT04354428

Document Date:

Version 2.1; 17 August 2020

About this Model Protocol

This model document is a protocol template created to align institutions engaged in evaluating hydroxychloroquine (HCQ), HCQ in combination with azithromycin, lopinavir-ritonavir (LPV/r), and other therapies for SARS-CoV-2 treatment. It contains the following:

- Sections written in black text are mandatory and must be consistent across the participating institutions to maintain scientific integrity of the project
- *Sections written in italics are optional and may be omitted as required (e.g., based on institutional needs and practices)*
- A site-specific protocol addendum template (Appendix 4) is provided to capture information that is specific to a given study site

CLINICAL STUDY PROTOCOL

Protocol Title:

Efficacy of Novel Agents for Treatment of SARS-CoV-2 Infection Among High-Risk Outpatient Adults: An Adaptive Randomized Platform Trial

Compound:

A. Hydroxychloroquine (terminated)
B. Azithromycin (terminated)
C. Lopinavir-ritonavir
D. Ascorbic acid (control)
E. Folic acid (control)

Short Title:

Treatment for SARS-CoV-2 in High-Risk Adult Outpatients

Protocol Version and Approval Date:

Version 1.4; 11 April 2020

Version 2.0; July 29, 2020

Version 2.1; August 17, 2020

Protocol Registry Number:

NCT04354428

Regulatory Agency Identifying Number(s):

Not applicable

Sponsor Name and Legal Registered Address:

University of Washington
4333 Brooklyn Ave NE
Box 359472
Seattle, WA 98195-9472

Funding Agency and Legal Registered Address:

Bill & Melinda Gates Foundation
1300 I St NW, Washington, DC 20005

Principal Investigator:

Dr. Christine Johnston, University of Washington
Phone: +1 206 520 4318; Email: cjohnsto@uw.edu

Sponsor Signatory Name and Title:

Signature	**Date**

Medical Monitor name and contact information is provided separately for each site.

Table of Contents

List of Tables

1. Synopsis

Narrow Title:

Efficacy of Treatment of SARS-CoV-2 Infection Among High-Risk Outpatient Adults: A Platform Trial

Expanded Title (pending discussions of approach):

Efficacy of Novel Agents for Treatment of SARS-CoV-2 Infection Among High-Risk Outpatient Adults: An Adaptive Randomized Platform Trial

Short Title:

Treatment for SARS-CoV-2 in High-Risk Adult Outpatients

Rationale:

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection causes mild illness confined to the upper respiratory track in 80% of people but can cause severe lower respiratory tract infection (LRTI) in 20%, particularly among those in high-risk groups, defined by advanced age (>60 years) and presence of comorbidities (e.g., cardiopulmonary disease, renal disease, diabetes mellitus). Progression to LRTI appears to frequently result in hospitalization for supplemental oxygen therapy and may lead to need for ventilator respiratory support and ultimately death. Given the rapid spread of the SARS-CoV-2 pandemic, interventions that avert adverse patient outcomes and reduce the strain on the medical system are urgently needed. In addition, prolonged viral shedding has been noted after infection; therapeutic strategies that can effectively reduce viral shedding, and potentially onward transmission, have the potential to shift the trajectory of the pandemic.

There are no FDA-approved therapeutic agents for any human coronavirus disease and many scientists and researchers have proposed potential agents based on *in vitro* and pre-clinical studies. These agents have potential benefits but also have inherent risks and limitations (e.g., scalability of supply). There is urgent need for controlled clinical studies in relevant patient populations to guide both policy makers and other clinical trialists on prioritized drugs, including a path toward future evaluations as data becomes available. This study intends to provide quick and informative data to assist in early decision-making and/or support combined analysis with other concurrently run studies.

Hydroxychloroquine (HCQ) is currently approved for the suppressive treatment (for extrahepatic phase of malaria) and treatment of acute attacks of malaria due to several *Plasmodium* strains. It is also indicated for the treatment of discoid and systemic lupus erythematosus and rheumatoid arthritis. With the first Food and Drug Administration approval in 1955, safety and tolerability of HCQ are well described. *In vitro*, HCQ displays antiviral activity against coronaviruses, including SARS-CoV-2. Pharmacologic modeling based on observed drug concentrations and *in vitro* drug testing suggest that treatment at approved doses (200 to 800 mg daily) could achieve levels in respiratory tract tissues to inhibit virus replication and shedding, potentially reducing adverse outcomes of infection and onward transmission. Preliminary, uncontrolled

studies suggest that HCQ could be clinically beneficial in persons with hospitalized coronavirus disease (COVID-19) and reduce viral shedding. The first arm of this platform trial explored HCQ as a treatment for early COVID-19.

Azithromycin is a broad-spectrum azalide antibiotic that was approved over 30 years for the treatment of a variety of bacterial infections, including pneumonia. Azithromycin has shown antiviral activity in vitro against Ebola, Zika, rhinovirus, and other respiratory viruses (Gautret 2020; Menzel 2016). The rationale for studying the combination of HCQ + azithromycin was early evidence of enhanced antiviral activity compared with HCQ alone in patients with COVID-19 disease in a small observational series reported by Gautret (2020). The second arm of this platform trial explored HCQ + azithromycin as a treatment for early COVID-19.

High dose lopinavir-ritonavir (LPV/r), a combination medication that has been used to treat human immunodeficiency virus/acquired immunodeficiency syndrome, also has shown *in vitro* activity against SARS (Chu 2004; Chen 2004; Wu 2004). Compared to ribavirin as a historical control group, it has been previously shown that LPV/r (400 mg and 100 mg, respectively) can reduce the risks of adverse clinical outcomes such as acute respiratory distress syndrome and death as well as viral load among patients with SARS (Chen 2004; Schoenfeld 2009; Chan 2003). A recent randomized clinical trial of standard-dose LPV/r utilized in hospitalized patients was not effective in decreasing time to clinical improvement or decreasing ribonucleic acid levels in respiratory secretions (Cao 2020). However, it is hypothesized that utilization of higher dose LPV/r earlier in infection may have improved efficacy at preventing LRTI. The third arm of this platform trial will investigate LPV/r as a treatment for early COVID-19.

This synopsis proposes a randomized platform trial for treatment of high-risk individuals in outpatient settings with SARS-CoV-2 infection, with primary outcomes focused on resolution of symptoms by Day 14 and reduction of upper respiratory viral shedding, defined as time to clearance (2 consecutive negative swabs).

A flexible platform trial design will allow additional candidate agents for SARS-CoV-2 infection that are identified and prioritized for testing to be incorporated into this protocol as additional arms. The design of the trial and the statistical analysis plan will allow seamless integration of new interventions. Some study procedures may be adapted based on a unique safety profile of these new candidates, but the core study elements will be maintained. If an intervention is shown to be effective, this design would also allow replacement of the placebo group with the effective intervention as the comparator.

Design:

Randomized (participant-blinded, treating clinician-blinded, laboratory-blinded, pharmacist-unblinded, and statistician-unblinded), multi-center, placebo-equivalent (ascorbic acid)-controlled, and blinded study of lopinavir/ritonavir for the treatment of SARS-CoV-2 infection in high-risk adults not requiring hospital admission. Additional arms will be added should new potential agents be discovered or combination treatments be proposed. In addition, arms may be dropped prior to completion if deemed futile or if there is a safety signal.

Population:

Men and women 18 to 80 years of age who had their first positive PCR test for SARS-CoV-2 within the past 72 hours and have COVID-19 symptoms (as per COVID-19 criteria).

High-risk cohort:

Eligible participants will be at increased risk of developing LRTI based on the established risk factors for severe COVID-19 disease (at least one of the following):

i. Age ≥60 years
ii. Presence of pulmonary disease, specifically moderate or severe persistent asthma, chronic obstructive pulmonary disease, pulmonary hypertension, emphysema
iii. Diabetes mellitus (type 1 or type 2), requiring oral medication or insulin for treatment
iv. Hypertension, requiring at least 1 oral medication for treatment
v. Immunocompromised status due to disease (e.g., those living with human immunodeficiency virus with a CD4 T-cell count of <200/mm^3)
vi. Immunocompromised status due to medication (e.g., persons taking 20 mg or more of prednisone equivalents a day, anti-inflammatory monoclonal antibody therapies, or cancer therapies)
vii. Body mass index ≥35 kg/m^2 (self-reported)

Low-risk cohort:

Eligible participants will be 18 to 59 years of age, inclusive, without any of the risk factors for developing severe COVID-19 disease (see high-risk cohort above).

COVID-19 symptoms criteria are as follows: At least TWO of the following symptoms: Fever (≥ 38ºC), chills, rigors, myalgia, headache, sore throat, new olfactory and taste disorder(s), OR At least ONE of the following symptoms: cough, shortness of breath or difficulty breathing.

Participants will be counseled about the preliminary *in vitro* data on LPV/r activity against SARS-CoV-2 and equipoise regarding efficacy in humans, given that there are only limited data at this time.

Interventions:

- Intervention A: HCQ (terminated)
- Intervention B: HCQ + azithromycin (terminated)
- Intervention C: LPV/r

Treatment Regimens:

- Treatment arm 3: LPV/r 800 mg-200 mg orally twice on Day 1, followed by 400 mg-100 mg orally twice daily for an additional 9 days (Days 2 to 10)
- Treatment arm 4: Ascorbic acid 1 gm orally twice on Day 1, followed by 500 mg orally twice daily for 9 days (Days 2 to 10)

Objectives and Endpoints:

Objectives	Endpoints
Primary	
• To test the efficacy of LPV/r compared to placebo to resolve COVID-19 symptoms within 14 days.	• COVID-19 symptoms are based on the following criteria: o At least TWO of the following symptoms: Fever (≥ 38ºC), chills, rigors, myalgia, headache, sore throat, new olfactory and taste disorder(s), OR o At least ONE of the following symptoms: cough, shortness of breath or difficulty breathing, OR o Severe respiratory illness with at least 1 of the following: ▪ Clinical or radiological evidence of pneumonia, OR ▪ Acute respiratory distress syndrome (ARDS), OR ▪ LRTI, defined by resting SpO2<93% sustained for 2 readings 2 hours apart AND presence of subjective dyspnea or cough Death or COVID-19-related hospitalizations will count as a failure to resolve symptoms.
• To test the efficacy of LPV/r compared to placebo to reduce SARS-CoV-2 viral shedding	• Time to clearance of nasal SARS-CoV-2, defined as 2 consecutive negative swabs (swabs collected daily Day 1 through Day 14) *Trial is statistically powered for this endpoint in both the high- and lower-risk populations, analyzed separately.*
Secondary	
• To test the safety of LPV/r compared to placebo for treatment of high-risk outpatients with SARS-CoV-2 infection	• Serious adverse events (including death and COVID-19-related hospitalization) and adverse events resulting in treatment discontinuation

• To test whether LPV/r has an effect on disease severity compared to placebo	• Peak score on WHO Ordinal Scale for Clinical Improvement • Peak score on modified Flu-PRO within the first 14 days
• To test whether LPV/r is associated with decreased viral shedding from self-collected nasal swabs over 14 days compared to placebo	• Proportion of days with SARS-CoV-2 detected from mid-nasal swabs by PCR • Median quantity of SARS-CoV-2 detected from mid-nasal swabs by PCR
Exploratory	
• To assess pharmacokinetics and exposure-response relationship of LPV/r	• LPV/r blood concentration in DBS

COVID-19: coronavirus disease; DBS: dried blood spot; Flu-PRO: inFLUenza patient-reported outcome; LPV/r: lopinavir-ritonavir; LRTI: lower respiratory tract infection; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; SpO2: blood oxygen saturation level; WHO: World Health Organization.

Sample Size:

The study will enroll 173 participants per arm to achieve 80% statistical power at 2-sided type I error rate of 5% for a pairwise comparison against the control to detect a treatment effect of hazard ratio of 1.5 for the primary outcome(s) assuming control event rate of 60% for the primary outcome and 5% dropout rate. Additional controls will be added to allow for contemporaneous controls for the LPV/r arm. Eligible participants will be randomized at a 1:1 ratio LPV/r + and Ascorbic acid. If a second eligible patient is in the same household, both will be assigned to the same regimen. A blinded sample size re-assessment may be done to possibly increase the target sample size, conditional on the observed incidence rate and within household correlation. Should a new intervention be added, the randomization ratio may be adjusted to favor the new arm, ensuring that control arm participants are still enrolled concurrently with active arms; an empirical Bayesian information borrowing method may be used to incorporate past control arm data for the comparison of new intervention that is added later.

Duration:

Twenty-eight days of clinical follow-up per participant.

Proposed Sites (up to 6): University of Washington, Tulane, Boston University, Cook County, and others to be discussed

2. Schedule of Activities

Procedure	Screening	Self-quarantine Treatment Period									EOS
	Day 0[a]	Day 1[a]	Day 2[a]	Day 3	Day 4[a]	Days 5 to 8	Day 9[a]	Days 10 to 13	Day 14[a]	Day 21	Day 28[b]
Informed consent	X										
Demography	X										
Sign HIPAA form	X										
Past and current medical conditions, including known pregnancy and/or lactation status	X										
Concomitant medications	X										
Laboratory documentation of COVID-19 testing	X										
Inclusion and exclusion criteria	X										
Randomization	X										
Mid-nasal swab		X	X	X	X	X	X	X	X	X	X
Study therapy (LPV/r, ascorbic acid)		##	#	#	#	#	#	#[c]			
Symptomatic relief medications as needed only (ondansetron, loperamide)		X	X	X	X	X	X	X			
Daily Survey (including dosing and swab adherence, concomitant medications)		X	X	X	X	X	X	X	X	X	
Modified Flu-PRO		X	X	X	X	X	X	X	X	X	X
WHO Ordinal Scale for Clinical Improvement									X	X	X
Participant collected daily vitals (including temperature, pulse, SpO2, respiratory rate)		X	X	X		X	X	X	X	X	X
QT monitoring*		X	X	X	X	X	X	X[d]			
Contact with study clinician or staff		X	X	X[e]	X	X[e]	X	X[e]	X	X[e]	X
Exit Contact Survey (including concomitant medications, symptoms, etc.)											X
Adverse event review		X	X	X	X	X	X	X	X	X	X
If in sub-study: dried blood spot sample for LPV/r concentration and anti-SARS-CoV-2 antibodies		*X*	*1 to 5 samples (no more than 2 per day) after dosing start*								*X*

Procedure	Screening	Self-quarantine / Treatment Period									EOS
	Day 0[a]	Day 1[a]	Day 2[a]	Day 3	Day 4[a]	Days 5 to 8	Day 9[a]	Days 10 to 13	Day 14[a]	Day 21	Day 28[b]

COVID-19: coronavirus disease; EOS: end of study; Flu-PRO: inFLUenza patient-reported outcome; HCQ: hydroxychloroquine; HIPAA: Health Insurance Portability and Accountability Act of 1996; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; SpO2: oxygen saturation; WHO: World Health Organization.

Note: ## 800 mg-200 mg LPV/r twice a day or placebo (ascorbic acid 1 gm) twice a day.

Note: # denotes 400 mg-100 mg LPV/r twice a day +placebo (ascorbic aid 500 mg) twice a day

[a] Screening and Day 1, 2, 4, 9, and 14 evaluations will be conducted through a web-based screening tool, HIPAA-compliant video conference (Telehealth), telephone, or text messaging. Screening (Day 0) and Day 1 evaluations may occur on the same day.

[b] For patients who are hospitalized on Day 28, the last visit will be ±3 days after discharge. The exit survey may also be conducted before day 28 if a participant terminates early and is willing.

[c] Last day of study medication will be administered on Day 10 LPV/r, and ascorbic acid.

[d] Last day of QT monitoring will be Day 11.

[e] These evaluations will be as needed/requested by study participant.

*May be omitted if not standard of care to monitor QTc for regimen

3. Introduction

This is a randomized, multi-center, placebo-equivalent (ascorbic acid + folic acid)-controlled, blinded platform trial for the treatment of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection in high-risk adults not requiring hospital admission. The trial will start with hydroxychloroquine (HCQ) (Intervention A), HCQ + azithromycin (Intervention B), and lopinavir-ritonavir (LPV/r) (Intervention C) as experimental interventions. Additional arms will be added as new agents or combinations are prioritized. In addition, if other agents are found to be effective, HCQ, HCQ + azithromycin, or LPV/r may become the control group against which new interventions are measured. Evaluations include safety and tolerability, SARS-CoV-2 viral shedding, and development of lower respiratory tract infection (LRTI). There will be 4 study arms (Arm 1: HCQ + placebo [folic acid], Arm 2: HCQ + azithromycin, Arm 3: LPV/r and Arm 4: placebo [ascorbic acid +/- folic acid],) enrolled to assess a daily dosing regimen administered for 10 days to prevent LRTI or decrease the time to symptom resolution and decrease viral shedding.

Up to 173 eligible adults (18 to 80 years of age) per arm will be randomized (at the level of household) in a 1:1 ratio to receive one of the following therapies:

- LPV/r 800 mg-200 mg orally twice on Day 1, followed by 400 mg-100 mg orally twice daily for an additional 9 days (Days 2 to 10)
- Placebo: Ascorbic acid 1 gm orally twice on Day 1, followed by 500 mg orally twice daily for 9 days

LPV/r will not appear similar to ascorbic acid. If there are more than 1 participant in the same household, all will be assigned to the same randomized group, preserving some degree of masking.

During the study, participants will perform the following:

- Collect mid-nasal swabs for viral detection for the co-primary trial endpoint
- Complete daily physical assessments for symptoms of LRTI and measurement of temperature, respiratory rate, pulse, and oxygen saturation (SpO2), QT monitoring (as needed), respiratory rate
- Complete up to 5 dried blood spots
- Complete surveys that will include questions about symptoms from both the drug regimen and respiratory and systemic symptoms, review of concomitant medications, and other pertinent topics

During the 28 study days, participants will take the medication, complete surveys, collect mid-nasal swab for viral quantification, and assess symptoms for progression to LRTI. Physical assessments will include daily temperature, SpO2, pulse, QT monitoring (as needed), dried blood spot and respiratory rate.

3.1. *Background*

SARS-CoV-2 is a coronavirus novel to the human population discovered in December 2019; it is currently the cause of a global pandemic (Huang 2020; Zhu 2020; Chen 2020). The World Health Organization (WHO) named the novel coronavirus SARS-CoV-2 and the disease caused by the virus as COVID-19.

As of 25 March 2020, nearly 500,000 persons were infected, with over 21,000 deaths reported from around the globe. As of August 17, there were over 21.6 million confirmed cases, with nearly 775,000 deaths. Person-to-person transmission has occurred across the globe. Accurate reporting is limited by availability of diagnostic testing. WHO declared the COVID-19 pandemic a Public Health Emergency of International Concern on 30 January 2020 (WHO News Release Jan 2020), and the United States declared a national emergency on 13 March 2020 (White House Press Release 2020).

Most deaths and severe pneumonitis have occurred in the elderly or in persons with underlying pulmonary or cardiac comorbidities or diabetes. In healthy adults, including pregnant women, it can cause a febrile, self-limited pneumonia. Infection appears less symptomatic in children and younger adults (Cai 2020). Nevertheless, the burden of this pandemic to the global health and economic systems is expected to be substantial. No acquired immunity to this novel viral infection appears to exist in the human population globally, and no effective treatment or preventative agent is licensed at this time.

As with many infectious epidemics, household contacts, first responders, caregivers, and medical personnel attending persons with COVID-19 are at high risk of infection. The incubation time requires 14 days of quarantine for exposed individuals not wearing personal protective equipment (Linton 2020), and on 03 March 2020, WHO declared a global shortage of personal protective equipment, leaving doctors, nurses, and other frontline workers dangerously ill-equipped to care for COVID-19 patients (WHO News Release Mar 2020). Extensive absences from the care network and health system will degrade the ability to care not only for those with COVID-19 but also for routine healthcare issues as well. At the height of local epidemic, the health care system becomes overburdened with patients with respiratory illness. To date, rigorous self-isolation and lockdown have been required to contain the SARS-CoV-2, leaving entire societies to abruptly stop normal life. Interventions are urgently needed to stop viral spread and to decrease the morbidity and mortality cause by the infection. The ability to stop viral replication to prevent transmission of the virus and to prevent disease progression, which is associated with decreased symptom duration , will be of benefit to the individual, and the health of the public. In addition, treating those at highest risk of disease progression and hospitalization will have the greatest impact on the pandemic.

3.2. *Study Rationale*

3.2.1. *COVID-19 and Antiviral Approaches*

SARS-CoV-2 is a novel betacoronavirus of zoonotic origin, similar to the coronaviruses severe acute respiratory syndrome coronavirus (SARS-CoV) and Middle East respiratory syndrome coronavirus (MERS-CoV). Based on current evidence, case fatality rate for SARS-CoV-2 is about 3%, which is significantly lower than that of SARS-CoV (10%) and MERS-CoV (40%)

(Chen 2020b). However, SARS-CoV-2 has potentially higher transmissibility (R0: 1.4 to 5.5) than both SARS-CoV (R0: 2 to 5) and MERS-CoV (R0: <1).

Our understanding of the viral pathogenesis of SARS-CoV-2 remains limited. However, it appears that the virus cell entry depends on the binding of the viral spike (S) proteins to cellular receptors and on S protein priming by host cell proteases. SARS-CoV-2, like SARS-CoV, uses the same receptor angiotensin converting enzyme 2 (ACE2) on pulmonary epithelial cells for entry and the transmembrane serine protease 2 for S protein priming (Hoffmann 2020). The receptor binding domain of lineage B betacoronaviruses is a single, continuous domain that contains all of the structural information necessary to interact with the host receptor. Fusion is mediated at the cell membrane, delivering the viral nucleocapsid inside the cell for subsequent replication. ACE2 expression is found in the lung epithelial cells, vascular endothelium, renal tubular epithelium, and epithelia of the small intestine. Viral shedding has been localized primarily to respiratory droplets and fecal samples (Zhu 2020).

Medications to treat and/or prevent SARS-CoV-2 need to inhibit aspects of the viral life cycle, ultimately blocking replication. Already-approved and available medications are ideal for immediate evaluation for SARS-CoV-2 infection treatment and prevention. Two potential targets for anti-SARS-CoV-2 medications are viral polymerases and proteases (Li 2019). Pilot clinical studies are already ongoing for SARS-CoV-2 using various repurposed antiviral medicines (https://www.who.int/emergencies/diseases/novel-coronavirus-2019/global-research-on-novel-coronavirus-2019-ncov). Similarities between SARS-CoV-2 with SARS-CoV and MERS suggest that antivirals with *in vitro* efficacy against SARS-CoV and MERS may be promising agents as SARS-CoV-2 post-exposure prophylaxis (PEP) (Li 2019; Lee 2019).

3.2.2. *Antiviral Effects of Chloroquine Analogues Against COVID-19*

Chloroquine (CQ) was discovered in 1934 by Bayer and was used in 1945 as an antimalarial, which became one of the most prescribed drugs globally, prior to the emergence of widespread drug resistance in *Plasmodium falciparum* (Slater 1993). CQ was found to be effective against rheumatoid tenosynovitis in 1951 (Mackenzie 1970). HCQ was licensed in the United States in 1955 as an antimalarial and as a drug for rheumatoid arthritis, and it was widely marketed for the latter due to a favorable safety profile with chronic use (Shippey 2018). The mechanisms of action for HCQ for treatment of rheumatoid arthritis and other autoimmune diseases are still not fully understood despite widespread use over the past 60 years (Schrezenmeier 2020).

CQ and HCQ have been proposed as potential agents for treatment and prevention against other infectious agents beyond malaria (Rolain 2007; Savarino 2015). The mechanism of action differs according to the pathogen: against intracellular bacteria and fungi by alkalinizing vacuoles containing the microorganisms, restoring the activity of other antibiotics, and against viral replication through alkalization of acidic organelles, namely, endosomes, lysosomes, and Golgi vesicles.

CQ is effective *in vitro* against SARS-CoV coronavirus in Vero E6 cells with a half-maximal effective concentration (EC_{50}) of ~8 μM (Keyaerts 2004) and had shown evidence of prevention activity *in vivo* (Vincent 2005). Hence, these re-purposed drugs were obvious hits for testing against SARS-CoV-2. *In vitro* inhibition in Vero E6 cells against the novel coronavirus, SARS-

CoV-2, has been published in recent weeks. Wang et al (2020) showed that the EC_{50} and EC_{90} for CQ in Vero E6 cells are 1.13 and 6.90 μM, respectively. Yao et al (2020) showed that the EC_{50} for CQ treatment of infected cells at 48 hours was 5.47 μM, whereas HCQ appeared slightly more potent, with EC_{50} of 0.72 μM at 48 hours. These levels appear to be within the range of exposures that could be achieved with standard HCQ treatment, and likely prophylaxis, due to concentrations of the drug achieved in the lung tissue. No *in vitro* data in the lung epithelial cells or any animal model data are available.

Multiple observational and small Investigator-initiated COVID-19 pneumonia treatment trials using CQ, HCQ, and variety of other medications are ongoing in China (https://www.who.int/emergencies/diseases/novel-coronavirus-2019/global-research-on-novel-coronavirus-2019-ncov). Gao et al (2020) reported anecdotal efficacy of CQ as treatment for COVID-19-associated pneumonia.

3.2.3. *Rationale for Drug Selection of HCQ*

The trial will test a daily regimen of HCQ for 10 days for treatment of SARS-CoV-2 (package insert, Appendix 5). Daily dosing has the highest likelihood to achieve sustained required drug levels for viral inhibition, as shown in the physiologically based pharmacokinetics (PBPK) modeling (Appendix 10). HCQ is commonly used daily in doses up to 600 mg of HCQ sulfate (465 mg base) per day for rheumatoid arthritis or systemic lupus erythematosus initially, with a usual maintenance dose of 200 mg (155 mg base) for maintenance therapy. HCQ and CQ are both commonly used in a weekly dosing schedule for malaria chemoprophylaxis.

HCQ is associated with a better safety profile for daily and chronic use than CQ, including 5 decades of experience with use in these dose ranges in adults and the elderly. It is on the WHO Essential Medicines List for use in rheumatic disorders and is widely prescribed as an anti-inflammatory for rheumatoid arthritis, systemic lupus erythematosus, and other autoimmune syndromes. Based on the limited *in vitro* data available, HCQ appears to be slightly more potent than CQ against SARS-CoV-2 (Yao 2020).

As the COVID-19 epidemic remains very fluid and new data are emerging from observational and clinical trials daily, this protocol is written to allow adaptation to incorporate additional medications beyond HCQ and HCQ + azithromycin.

3.2.4. *Rationale for Dosing Schedule of HCQ*

HCQ is a long-acting drug with a terminal half-life of approximately 40 days. It is well absorbed, moderately protein bound, and will accumulate in tissues including the lung, heart, liver, and kidneys. It is typically given with a loading dose of approximately 2-fold the standard dose to accelerate achieving steady-state drug concentrations (Furst 1999). Since the drug will be used in those with SARS-CoV-2 upper respiratory tract infection, it is desirable to quickly achieve adequate drug levels to decrease viral replication.

A PBPK model was built (SIMCYP simulator version 18) using physical and chemical parameters of HCQ obtained from the literature (Collins 2018); pharmacokinetic (PK) parameters (liver intrinsic clearance, fa, ka) were determined from clinical data (Tett 2018). This PBPK model was used to simulate HCQ concentrations in plasma and lung fluid following 5 proposed dosing regimens in order to select an optimal regimen for the Peking University

Third Hospital's ongoing trial of HCQ in China. The combination of *in vitro* antiviral concentration-effect and predicted drug concentrations in this study were used to propose a loading dose of 400 mg HCQ twice on Day 1, and drug levels will remain above the EC_{50} for at least 14 days of treatment and potentially beyond Day 21.

A second study (BYSY-DCTC-CPPO-HCQ-PBPKAR) was undertaken to simulate HCQ concentration-time profiles in plasma, whole blood, and lung. Since elderly patients have reduced glomerular filtration rate (GFR), simulations were conducted using a healthy Caucasian healthy population with renal injury (GFR 30 to 60 mL/min) and compared to a population with normal renal function to support the study design of therapeutic use of HCQ.

This protocol will investigate a single dosage of HCQ. Participants will receive a loading dose of 400 mg twice on Day 1, followed by 200 mg twice a day for 9 additional days. Subsequent investigations will be encouraged to undertake a more rigorous exposure-response assessment to define optimal dosing, including exploration of the lowest possible effective dose, and possible alternate dosing schedules (i.e., weekly instead of daily).

3.2.5. *Antiviral Effects of Azithromycin Against COVID-19*

Azithromycin is a broad-spectrum azalide antibiotic used to treat a number of bacterial infections, including pneumonia. Azithromycin has shown antiviral activity *in vitro* against Zika, Ebola, rhinoviruses, and other respiratory viruses (Gautret 2020; Menzel 2016). Although the mechanism of its antiviral activity is not clear, some findings suggest it may be associated with augmentation of interferon response (Menzel 2016). Alternatively, azithromycin may convey antiviral activity by increasing the pH of cell organelles such as endosomes and the *trans*-Golgi network (Poschet 2020). Changing the pH of intracellular vesicles may alter the glycosylation of ACE2, a key receptor for cell entry for SARS-CoV-2 (Belouzard 2012; Wrapp 2020).

3.2.6. *Rationale for Drug Selection of Azithromycin*

The trial will test a daily regimen of azithromycin in combination with HCQ in the treatment of SARS-CoV-2 (Zithromax package insert, Appendix 6). In the Gautret study (Gautret 2020), among 20 participants with confirmed COVID-19 who were treated with HCQ, 6 received azithromycin (500 mg on Day 1 followed by 250 mg/day on Days 2 to 5) to prevent bacterial pneumonia superinfection. On Day 6, all participants who were treated with the HCQ + azithromycin combination showed virologic cure as measured by PCR of nasopharyngeal samples compared with 57.1% of participants treated with HCQ alone.

Azithromycin, which has been approved for use for >30 years, is associated with a favorable safety profile. It is on the WHO's List of Essential Medicines, and WHO classifies it as critically important for human medicine. Azithromycin is widely available worldwide. Common short-term drug toxicities include nausea, vomiting, diarrhea and upset stomach.

3.2.7. *Rationale for Dose Selection of Azithromycin*

The dose of azithromycin selected for this study aligns with the Dosage and Administration recommendations in the United States prescribing information (USPI) for the treatment of community-acquired pneumonia. This regimen is 500 mg as a single dose on Day 1, followed by 250 mg once daily on Days 2 through 5. Azithromycin does not undergo significant metabolism and does not complex with or induce cytochrome P450 (CYP), thus minimizing the potential for drug interactions. Because azithromycin has a long terminal half-life (79 hours with 500 mg oral dosing), once daily or single-dose treatments are possible (Ballow 1992).

The dose regimen of 500 mg as a single dose on Day 1, followed by 250 mg once daily on Days 2 through 5 was used in the recently published study of HCQ and azithromycin in patients with COVID-19 (Gautret 2020). Using a PBPK model, azithromycin lung concentrations were simulated for this dose regimen to ensure that physiologically relevant lung exposure can be reached. Results are presented in Appendix 8. Based on the simulations, it appears that a dosing regimen of 500 mg for 1 day followed by 250 mg once daily for 4 days is able to attain concentrations of azithromycin in the lung that would approach the in vitro EC_{50} value reported for inhibition of SARS-CoV-2 by azithromycin (Touret 2020).

3.2.8. *Antiviral Effects of Lopinavir and Ritonavir Analogues Against COVID-19*

LPV/r, also known as Kaletra, is a combination medication that has been used to treat human immunodeficiency virus (HIV)/acquired immunodeficiency syndrome. Lopinavir is a HIV type 1 aspartate protease inhibitor that is usually "boosted" with ritonavir to increase the plasma half-life of lopinavir through the inhibition of CYP. Lopinavir has been shown to have *in-vitro* inhibitory activity against SARS-CoV (Chu 2004; Chen 2004; Wu 2004). Compared to ribavirin as a historical control group, it has been previously shown that LPV/r (400 mg and 100 mg, respectively) can reduce the risks of adverse clinical outcomes such as acute respiratory distress syndrome and death as well as viral load among patients with SARS (Chen 2004; Schoenfeld 2009; Chan 2003). Based on *in vitro* testing and previous clinical evaluation, LPV/r is an important treatment regimen that should be tested for SARS-CoV-2. Although a recent open-label clinical trial did not show efficacy of LPV/r to decrease days of hospitalization or viral shedding, this trial focused on patients who were already hospitalized, likely due to LRTI and used a suboptimal dosing strategy (Cao 2020). The proposed trial will treat participants when they still have upper respiratory tract infection, earlier in the illness, and will use a higher loading dose, which will allow the LPV/r to reach a plasma concentration that exceeds the EC_{50} for SARS-CoV-2.

3.2.9. *Rationale for Drug Selection of LPV/r*

The trial will test a daily regimen of LPV/r for 10 days for treatment of SARS-CoV-2 (package insert, Appendix 7). Daily dosing after a large loading dose has the highest likelihood to achieve sustained required drug levels for viral inhibition. LPV/r is commonly used daily in regimens of combination antiretroviral therapy for the treatment of HIV infection.

LPV/r is associated with a favorable safety profile. Short-term drug toxicities include moderate nausea, diarrhea, and liver function test abnormalities. It is on the WHO Essential Medicines List for use in HIV infection, and is widely available throughout the world.

3.2.10. *Rationale for Dose Selection of LPV/r*

To obtain adequate levels to exceed the 50% inhibitory concentration for SARS-CoV-2, LPV/r will be dosed at 800 mg-200 mg twice daily on the first day. The dose will be decreased to 400 mg-100 mg twice daily (United States Food and Drug Administration [FDA]-approved dose) for the remainder of the clinical trial.

3.2.11. *Rationale for Ascorbic Acid Control as a Comparator for HCQ and LPV/r*

In healthy adults, COVID-19 disease is likely to present as an upper respiratory viral infection, characterized by a febrile disease with cough and fatigue (Huang 2020; WHO Feb 2020). Symptom reporting may vary based on the participants' perception as to whether they are taking HCQ or ascorbic acid (Barrett 2011), but the primary study endpoints are not affected. There is no rigorously proven therapy for individuals with outpatient COVID-19 disease, although multiple therapies are under investigation.

Because there is no established therapy, use of a control is acceptable and ethical for both the participants' health and safety as well as ensuring the most rigorous trial design to evaluate an intervention for COVID-19 disease caused by SARS-CoV-2. Participants will be blinded to their allocation to the extent possible. However, LPV/r will not appear similar to ascorbic acid. If >1 individual from the same household is enrolled, all will be assigned to the same randomized group.

The dose of ascorbic acid chosen for this protocol is safe. All participants, regardless of assigned group, will be able to take additional ascorbic acid (e.g., over the counter vitamins or through food) should they choose, as there is no known maximum daily safe dose of ascorbic acid. Clinical trial evidence has demonstrated that ascorbic acid, alone or in combination with other micronutrients, does not substantially reduce the risk of upper respiratory infections or severe consequences of infectious processes (Fujii 2020; Graat 2002); thus, ascorbic acid is not expected to have a prevention effect for SARS-CoV-19 and is considered a placebo-equivalent product for this study.

3.2.12. *Rationale for Folic Acid Control as a Comparator for Azithromycin*

Folic acid, also known as vitamin B9 is one of the B vitamins and commonly used as a dietary supplement. Specifically, folic acid is also used as a supplement during pregnancy to reduce the risk of neural tube defects in the baby. Folic acid is on the WHO's List of Essential Medicines, widely available worldwide, and inexpensive.

Folic acid has no known antiviral activity. The risk of toxicity from folic acid is low, because folic acid is water-soluble and excreted through urine. The 100% daily recommended for folic acid is 400 µg, and the upper intake level is established as 1000 µg/day. No health risks have been associated with high intake of folic acid from food sources.

As folic acid is not expected to have a preventative effect for SARS-CoV-19 it is considered an acceptable placebo-equivalent product for this study.

3.3. *Benefit/Risk Assessment*

COVID-19 disease can be unpredictable in its severity, but a 3.4% mortality rate has been observed among clinical pneumonia cases. The elderly (>60 years of age) and those with medical comorbidities (e.g., cardiopulmonary disease, renal disease, diabetes mellitus) are at highest risk of poor outcomes (Huang 2020; Zhu 2020; Chen 2020). Moreover, transmission in younger persons amplifies infection in communities, putting susceptible persons at risk. There is no proven drug for treatment of those with COVID-19 disease. HCQ and LPV/r have an excellent safety record at the proposed doses for many years and both agents have potent viral suppression *in vitro*. The safety profile of azithromycin at the proposed dose is similarly well characterized with safety considerations and monitoring guidance provided in the label. There is equipoise as to whether the *in vitro* efficacy of HCQ, HCQ + azithromycin, LPV/r, or any other drug or drug combination, will translate into efficacy to decrease symptom duration and decrease viral shedding. Thus, the potential benefit-to-risk ratio for testing HCQ, HCQ + azithromycin, and LPV/r as treatment is favorable in this population.

4. Objectives and Endpoints

Initial clinical endpoints included progression to lower respiratory tract infection, hospitalization, and death. The event rate for these endpoints was too low to achieve the study objective, and therefore the clinical endpoint was changed to symptom resolution within 14 days in Version 2.0.

Objectives	Endpoints
Primary	
• To test the efficacy of LPV/r compared to placebo to resolve COVID-19 symptoms within 14 days.	• COVID-19 symptoms are based on the following criteria: o At least TWO of the following symptoms: Fever (≥ 38ºC), chills, rigors, myalgia, headache, sore throat, new olfactory and taste disorder(s), OR o At least ONE of the following symptoms: cough, shortness of breath or difficulty breathing, OR o Severe respiratory illness with at least 1 of the following: ▪ Clinical or radiological evidence of pneumonia, OR ▪ Acute respiratory distress syndrome (ARDS), OR ▪ LRTI, defined by resting SpO2<93% sustained for 2 readings 2 hours apart AND presence of subjective dyspnea or cough Death or COVID-19-related hospitalizations will count as a failure to resolve symptoms.
• To test the efficacy of LPV/r compared to placebo to reduce SARS-CoV-2 viral shedding	• Time to clearance of nasal SARS-CoV-2, defined as 2 consecutive negative swabs (swabs collected daily Day 1 through Day 14)
Secondary	
• To test the safety of LPV/r compared to placebo for treatment of high-risk outpatients with SARS-CoV-2 infection	• Serious adverse events (including death and COVID-19-related hospitalization) and adverse events resulting in treatment discontinuation

• To test whether LPV/r has an effect on the duration of hospitalization among persons who become hospitalized with COVID-19 disease	• Days of COVID-19-related hospitalization
• To test whether LPV/r has an effect on disease severity compared to placebo	• Peak score on WHO Ordinal Scale for Clinical Improvement • Peak score on modified Flu-PRO within the first 14 days
• To test whether LPV/r is associated with decreased viral shedding from self-collected nasal swabs over 14 days compared to placebo	• Proportion of days with SARS-CoV-2 detected from mid-nasal swabs by PCR • Median quantity of SARS-CoV-2 detected from mid-nasal swabs by PCR
Exploratory	
• To assess pharmacokinetics and exposure-response relationship of LPV/r	• LPV/r blood concentration in DBS

COVID-19: coronavirus disease; DBS: dried blood spot; Flu-PRO: inFLUenza patient-reported outcome; LPV/r: lopinavir-ritonavir; LRTI: lower respiratory tract infection; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; SpO2: blood oxygen saturation level; WHO: World Health Organization.

5. Study Design

5.1. *Overall Design*

The overarching goal of this study is to assess the effectiveness of interventions on the symptom duration, and length of viral shedding among adult outpatients with SARS-CoV-2 infection to inform public health control strategies.

This is a randomized, multi-center, placebo-equivalent (ascorbic acid +/- folic acid)-controlled, blinded platform trial. Stratified randomization based on different trial sites will be used. The trial started with HCQ (Intervention A) and HCQ + azithromycin (Intervention B) to assess the efficacy on the prevention of LRTI progression. With this protocol amendment, HCQ and HCQ/azithromycin were stopped, and LPV/r (Intervention C) was added. The trial may allow for additional agents to be added and tested against placebo with standardized eligibility criteria, outcomes, and measurements.

This study will enroll 173 eligible adults per arm (18 to 80 years of age) with high risk for disease progression or at low risk at baseline who are PCR-confirmed SARS-CoV-2 infection with symptoms compatible with COVID-19. Eligible participants will be enrolled and randomized in a 1:1 ratio to LPV/r or Vitamin C. Recruitment rate will be assessed on a weekly basis starting at the end of second week after the first eligible patient is recruited and randomized. Blinded sample size re-assessment will be done to increase the sample size target, should a lower control event rate (CER) and/or higher dropout rate than expected be observed. A decision for a possible interim analysis will be made during the trial based on the information on the observed recruitment rate, the CER, the dropout rate, and the final sample size target, if applicable. The decision for interim analysis will be made in a blinded manner (e.g., based on pooled number of events). The interim monitoring plan (written by the Study Statistician) will define monitoring bounds to maintain the 2-sided type I error rate at the desired 5% (e.g., 97.5% or higher probability of superiority over the control group). Should new experimental candidates be added during the trial, randomization ratios may be adapted to favor the new arms.

It is anticipated that additional treatment modalities will be incorporated into this protocol, in an adaptive fashion. Those will be added through later protocol amendments, which will define additional sample size needs and any potential change to randomization (e.g., changing the randomization ratio, continuation/discontinuation of any of the existing arms, etc.).

Participants will be counseled about the preliminary *in vitro* data on and LPV/r activity against SARS-CoV-2 and equipoise regarding efficacy in humans, given that there are only limited data from an uncontrolled study at this time.

Participants may participate in a sub-study where they will be asked to provide a dried blood spot (DBS) sample for therapy concentration and pharmacokinetics of the medications as well as for SARS-CoV-2 antibody testing.

An independent data and safety monitoring board (DSMB) will be convened for this study with expertise in COVID-19 or respiratory viruses and emerging epidemics as well as biostatistics. The purpose of the DSMB is to monitor the study for operational futility, social harms, and efficacy. The DSMB will also review the blinded sample size re-assessment plan and proposed

sample size changes and make recommendations on the randomization ratio, in case the new intervention arms are added.

This multi-center study will be conducted in high COVID-19 disease incidence areas in the United States, with the potential to expand enrollment to international sites.

5.2. *Participant and Study Completion*

Up to 173 high and low-risk participants per arm will be randomly assigned to study treatment or control. This sample size target may increase based on the blinded sample size re-assessment.

A participant is considered to have completed the study if he/she has completed all phases of the study, including the last scheduled procedure shown in the Schedule of Activities (SoA).

This study will be considered completed when sufficient number of participants complete the study to enable appropriate evaluation of the primary endpoint.

6. Study Population

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

6.1. *Inclusion Criteria*

Participants are eligible to be included in the study only if all of the following criteria apply (173 participants per arm):

High-risk cohort (:

1. Men or women 18 to 80 years of age, inclusive, at the time of signing the informed consent
2. Willing and able to provide informed consent
3. Laboratory confirmed SARS-CoV-2 infection, with first positive PCR test results within the past 72 hours
4. COVID-19 symptoms, based on the following criteria: At least TWO of the following symptoms: Fever (≥ 38ºC), chills, rigors, myalgia, headache, sore throat, new olfactory and taste disorder(s), OR o At least ONE of the following symptoms: cough, shortness of breath or difficulty breathing
5. Access to device and internet for Telehealth visits
6. At increased risk of developing severe COVID-19 disease (at least one of the following)
    a. Age ≥60 years
    b. Presence of pulmonary disease, specifically moderate or severe persistent asthma, chronic obstructive pulmonary disease, pulmonary hypertension, emphysema
    c. Diabetes mellitus (type 1 or type 2), requiring oral medication or insulin for treatment
    d. Hypertension, requiring at least 1 oral medication for treatment
    e. Immunocompromised status due to disease (e.g., those living with HIV with a CD4 T-cell count of <200/mm^3)
    f. Immunocompromised status due to medication (e.g., persons taking 20 mg or more of prednisone equivalents a day, anti-inflammatory monoclonal antibody therapies, or cancer therapies)
    g. Body mass index ≥35 (self-reported)

Low-risk cohort:

1. Men or women, 18 to 59 years of age, inclusive without any risk factors for developing severe COVID-19 disease (point 5 above)
2. Willing and able to provide informed consent
3. Laboratory confirmed SARS-CoV-2 infection, with first positive PCR test results within the past 72 hours
4. COVID-19 symptoms, based on the following criteria: At least TWO of the following symptoms: Fever (≥ 38ºC), chills, rigors, myalgia, headache, sore throat, new olfactory

and taste disorder(s), OR o At least ONE of the following symptoms: cough, shortness of breath or difficulty breathing

5. Access to device and internet for Telehealth visits

6.2. *Exclusion Criteria*

Participants are excluded from the study if any of the following criteria apply:

1. Known hypersensitivity to HCQ or other 4-aminoquinoline compounds*
2. Known hypersensitivity to azithromycin or other azalide or macrolide antibiotics*
3. Known hypersensitivity to LPV/r
4. Currently hospitalized due to COVID-19 and/or with discharge planning >24 hours out*
5. Signs of respiratory distress prior to randomization, including respiratory rate >24
6. Current medications include HCQ*, azithromycin*, or LPV/r
7. Current medications include a protease inhibitor, ritonavir, or cobicistat
8. Concomitant use of other anti-malarial treatment or chemoprophylaxis*
9. History of retinopathy of any etiology*
10. Psoriasis requiring immunosuppressive medications. Psoriasis managed with topical steroids may be permitted as per discretion of study clinician.*
11. Porphyria*
12. Chronic kidney disease (Stage IV or receiving dialysis)
13. Known bone marrow disorders with significant neutropenia (polymorphonuclear leukocytes <1500) or thrombocytopenia (<100 K)
14. Concomitant use of digoxin, cyclosporin, cimetidine, amiodarone, or tamoxifen*
15. Known cirrhosis
16. Known personal or family history of long QT syndrome
17. History of coronary artery disease with a history of graft or stent
18. History of heart failure, Class 2 or greater using the New York Heart Association functional class
19. Taking medications associated with prolonged QT and known risk of torsades de points (see Section 7.9.1). These medications may include some antipsychotic and antidepressant medications.
20. Taking warfarin (Coumadin or Jantoven)
21. Taking medications contraindicated with the use of LPV/r, including medications metabolized by cytochrome P450 3A (CYP3A), which may accumulate in patients receiving LPV/r (see Section 7.9.1). These medications may include anticoagulants, anticonvulsants, rifampin, antifungals, and immunosuppressants.
22. HIV infection with detectable plasma HIV ribonucleic acid (RNA) level or receiving protease inhibitors as part of combination antiretroviral therapy
23. Known history of glucose-6-phosphate-dehydrogenase deficiency*
24. History of myasthenia gravis*
25. Unwilling to use nonhormonal contraception such as additional barrier method during study participation when using hormonal contraception

*Exclusion criteria related to HCQ or azithromycin, will not be included as these arms have been discontinued

Note: Pregnant and lactating persons will be eligible for enrollment into this study. HCQ, azithromycin, and LPV/r have been shown to be safe in pregnant women.

6.3. *Screen Failures*

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently randomly assigned to study treatment. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the Consolidated Standards of Reporting Trials publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

Individuals who do not meet the criteria for participation in this study (screen failure) can be rescreened if there is a change in their eligibility.

6.4. *Recruitment*

Each site will establish local recruitment and screening methods that operationalize protocol-specified requirements for eligibility determination in a manner that is tailored to and most efficient for the local study setting and target study population. Each site will use a variety of recruitment approaches, including direct recruitment at clinics, referrals from other providers and SARS-CoV-2 testing sites and laboratories, and use of online and social networking websites and apps. Recruitment materials may educate participants about COVID-19, transmission within households, and epidemiology in the community.

If additional participants are required to increase sample size or add additional treatment arms, the number will be communicated to approval bodies for authorization.

6.5. *Co-enrollment Guidelines*

Participants may be co-enrolled in other research studies, provided that these are observational studies only. Any other exception requires approval of the Principal Investigators; if a participant clinically worsens, such as requiring hospitalization, it is expected that an exception will be automatically granted and participation in treatment studies permitted. The study team should be consulted for co-enrollment in studies that do not meet this guidance or if there are questions about eligibility for co-enrollment. For any co-enrolled study, combined blood draws should not exceed current Red Cross phlebotomy guidance.

7. Treatments

Study treatment is defined as any investigational treatment(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

7.1. *Treatments Administered*

Study Treatment	Hydroxychloro-quine sulfate	Ascorbic acid	Lopinavir-ritonavir	Ascorbic acid	Azithromycin	Folic acid	Ondansetron	Loperamide
Dosage Formulation	200 mg (155 mg base) tablets	250 mg tablets	200 mg-50 mg tablets	250 mg tablets	250 mg tablets	400 µg tablets	4 mg tab	2 mg tab
Route of Administration		Oral						
Dosing Instructions	Take 2 tablets twice on Day 1 and 1 tablet twice a day for the subsequent 9 days, for a total of 10 days of treatment. Take at approximately the same time of the day with a meal or a glass of milk.		Take 4 tablets every 12 hours on Day 1, and 2 tablets twice daily for the 9 subsequent days, for a total of 10 days of treatment. Take at approximately the same time each day with a meal.		Take 2 tablets once on Day 1 and 1 tablet once a day for the subsequent 4 days, for a total of 5 days of treatment. Tablets can be taken with or without food.		Take 1 tablet as needed for nausea every 8 hours. Do not exceed 8 pills per 24 hour period.	Take 2 tablets as needed for diarrhea. Take 1 additional pill with each episode of diarrhea. Do not exceed 4 pills per 24 hour period.
	If a dose is missed, it should be taken as soon as possible. If it is less than 4 hours before the next dose, the dose should be skipped.							
Packaging and Labeling	The medication for home delivery will be dispensed in an otherwise-unmarked container with the study label. The container will be labeled with a unique identifier. The container will be packed in a standard box used for mail delivery of medications as needed.							
Manufacturer	Sandoz	Cardinal Health	Abbvie		Greenstone LLC NDC 59762-2198-3 (blister pack) NDC 59762-2198-3 (bottle)	Leader (distributed by Cardinal Health)	TBD	TBD

7.2. *Risks to the Participants*

7.2.1. *Risks Associated Administration with HCQ*

With tens of millions of doses of HCQ administrated for malaria and autoimmune diseases, the side effect profile of HCQ is well described and the drug is generally well tolerated. With short-term administration (as opposed to chronic/year-long use in rheumatologic disease management), the major AEs are gastrointestinal (nausea, vomiting, dyspepsia, abdominal cramps, and diarrhea) and transient skin rashes. The gastrointestinal symptoms may vary by specific generic manufacturer of HCQ (Srinivasa 2017) and are best managed by taking the drug with food or a glass of milk (Appendix 5). A transient rash, most commonly morbilliform or psoriasiform, can develop in 10% of participants, often with a sustained loading dose, and is often managed by lowering the dose. To avoid this potential side effect, this protocol is using a short loading dose, not a sustained one. Uncommonly, idiosyncratic leukopenia/thrombocytopenia can occur and the drug should not be given to those with underlying bone marrow disorders. Lastly, hypoglycemia can occur and those taking insulin or glucose-lowering drugs are at risk; blood glucose should be monitored.

The safety of HCQ for treatment of patients with COVID-19 disease is unknown. COVID-19 may be associated with cardiac effects. HCQ may prolong QT, resulting in arrhythmias. Participants will be monitored for QT prolongation and counseled about this risk.

Long-term manifestations of HCQ, including retinitis, renal and hepatic disease, and cardiomyopathy (Appendix 5), are not likely in short-term exposure.

7.2.2. *Risks Associated with Azithromycin*

The most common adverse reactions to azithromycin are diarrhea, nausea, abdominal pain, or vomiting (Zithromax USPI, Appendix 6). Severe side effects include serious allergic reactions, including, but not limited to, anaphylaxis, angioedema, and dermatologic reactions such as Stevens-Johnson syndrome. Abnormal liver function, hepatitis, cholestatic jaundice, hepatic necrosis, and hepatic failure have been reported, some of which have resulted in death. Azithromycin can cause abnormal changes in the cardiac electrophysiology that may lead to a potentially fatal irregular heart rhythm. It may increase the risk of death, especially in those with heart problems, compared with those on other antibiotics such as amoxicillin or no antibiotic. The FDA warning indicated that people with pre-existing conditions are at particular risk, such as those with QT interval prolongation, proarrhythmic conditions, low blood levels of potassium or magnesium, a slower than normal heart rate, or those who use certain drugs to treat abnormal heart rhythms. Elderly patients may be more susceptible to development of torsades de pointes arrhythmias.

7.2.3. *QT Risks Associated with Coadministration of HCQ and Azithromycin*

Combination treatment with HCQ + azithromycin was associated with heart-rate corrected QT interval (QTc) prolongation in a study of 84 patients with SARS-CoV-2 (Chorin 2020). Maximal QTc prolongation was observed between Days 3 and 4 of treatment. QTc increase from baseline

was >40 ms in 30% of the patients and 11% exhibited severely prolonged QTc, defined as a maximum increase to >500 ms. Multivariate analysis showed that the development of acute renal failure rather than baseline QTc was a significant predictor of severe QTc prolongation in these patients.

7.2.4. *Risks Associated with LPV/r*

The safety and tolerability of LPV/r has been well characterized through clinical trials and postmarketing experience since first authorization for use in 2001 for the approved HIV indication with over 7 million patient years of exposure. The LPV/r prescribing information (Appendix 7) describes the known safety profile of LPV/r. Based on clinical trials and postmarketing experience, the most frequently reported adverse drug reactions among patients receiving LPV/r alone or in combination with other antiretroviral agents were gastrointestinal disorders (including diarrhea, nausea, vomiting, and upper and lower abdominal pain), fatigue/asthenia, respiratory tract infection (upper and lower), lipid elevations (hypercholesterolemia and hypertriglyceridemia), musculoskeletal pain (including arthralgia and back pain), and headache (including migraine). Key safety concerns include metabolic abnormalities such as dyslipidemia and insulin resistance, pancreatitis, hepatotoxicity and toxicity in preterm neonates of LPV/r oral solution. In addition, in the HIV population, immune reconstitution inflammatory syndrome manifesting as autoimmune disorders (such as Grave's disease) has been reported. Important potential risks include PR prolongation at therapeutic dosing, and QT prolongation with supratherapeutic doses. LPV/r interacts with several drugs since it is an inhibitor of the P450 isoform CYP3A and is likely to increase the plasma concentration of drugs that are metabolized by CYP3A4. Therefore, LPV/r should not be coadministered with drugs primarily metabolized by CYP3A and for which elevated plasma concentrations are associated with serious and/or life-threatening events. A list of such products is included in the LPV/r label. Rare reports of second- or third-degree atrioventricular block in patients with underlying structural heart disease and pre-existing conduction system abnormalities, or in patients receiving drugs known to prolong the PR interval such as verapamil, have been reported in patients receiving LPV/r. LPV/r should be used with caution in such patients. In addition, QT prolongation with supratherapeutic doses and when LPV/r is coadministered with drugs known to prolong the QT interval has been reported. Because LPV/r is principally metabolized by the liver, caution should be exercised when administering this drug to patients with impaired hepatic function. Extra monitoring is recommended when diarrhea occurs. The relatively high frequency of diarrhea during treatment with LPV/r may compromise the absorption and efficacy (due to decreased compliance) of LPV/r or other concurrent drugs. Serious persistent vomiting and/or diarrhea with LPV/r use might also compromise renal function. The safety of LPV/r for treatment of patients with COVID-19 disease is unknown. COVID-19 may be associated with cardiac effects. LPV/r may prolong QT at supratherapeutic doses and prolong PR at therapeutic doses, resulting in arrhythmias.

7.2.5. *Risks Associated with COVID-19 Diagnosis*

Enrollment in this protocol will not impact the public health department’s advice for self-quarantine. Enrollment may improve morale during quarantine for COVID-19 infection.

COVID-19 infection may be associated with anxiety, and the ability to monitor for disease progression and interact with study clinicians may allay anxiety.

7.3. *Strategies to Minimize Risk*

7.3.1. *Dose Selection*

The recommended dose of HCQ for chronic use in lupus erythematosus is 200 to 400 mg daily, a dose that is safely taken for years including among elderly patients (≥65 years). The selection of the time-limited dosing for this study is likely to be safe with transient adverse events (AEs) (gastrointestinal symptoms and rashes) that are self-remitting.

The recommended dose of azithromycin is 500 mg as a single dose on Day 1, followed by 250 mg once daily on Days 2 through 5. This dose has been safely used for several years for treatment of a wide variety of bacterial infections, including pneumonia.

The recommended dose of LPV/r is 400 mg-100 mg twice daily, a dose that has been utilized for several years for treatment of HIV infection as part of combination antiretroviral therapy. To rapidly obtain steady state in this trial, the medication will be dosed at a higher amount (double) on the first day of administration. The loading dose may be associated with GI side effects (nausea, vomiting, and diarrhea).

7.3.2. *Management of Participants to Limit Risks of SARS-CoV-2 Transmission*

To limit the transmission of SARS-CoV-2, participants will receive visits via secure Telehealth in order to limit the movement of persons with potential SARS-CoV-2 and leave clinical space free for ill patients requiring care. Also, to limit exposure in waiting rooms and pharmacies, clinical specimens will be self-collected and medications will be delivered to homes. This will also eliminate exposure of study personnel to SARS-CoV-2.

7.4. *Dose Modification and Toxicity Management*

If a study therapy dose is missed, it should be taken as soon as possible. If it is less than 4 hours before the next dose, the dose should be skipped.

Modification for toxicities is discussed below. Only toxicities related to study medications provided through the study will be considered in the toxicity management section.

LPV/R is known to cause GI side effects, specifically nausea, vomiting, abdominal pain, and diarrhea. Since participants will have difficulty obtaining medications for symptomatic relief due to these symptoms during the COVID-19 quarantine period, medications to control these symptoms will be given to each participant. Symptomatic medication will be included in separate medication vials and will clearly labeled for participants. Ondansetron 4 mg tabs (#10) and loperamide 2 mg tabs (#10) will be provided to each participant.

Grade 1 or 2

Participants who develop Grade 1 or 2 toxicity (per Division of Acquired Immunodeficiency Syndrome [DAIDS] AE Grading Table; see: https://rsc.tech-res.com/docs/default-

source/safety/division-of-aids-(daids)-table-for-grading-the-severity-of-adult-and-pediatric-adverse-events-corrected-v-2-1.pdf) that is considered by the site Investigator to be related to the study medication may continue study treatment at the discretion of the site Investigator with close follow up. If a participant chooses to discontinue study treatment, the site should notify the study protocol team within 7 days. These participants will remain on study, off study treatment, and have all evaluations performed.

Grade 3

- Participants who develop a Grade 3 symptomatic toxicity thought by the site Investigator to be related to study drug should have study product withheld, and the site should consult with the Core Protocol team. The participant should be reevaluated every 2 days until the AE returns to Grade ≤2, at which time study drug may be reintroduced at the discretion of the site Investigator in consultation with the protocol team.
- Participants experiencing Grade 3 toxicity requiring permanent discontinuation of study product should be followed up weekly until resolution of the toxicity. Participants will have premature study treatment discontinuation evaluations performed. These participants will remain on study, off study treatment, and have all evaluations performed per the SoA.

Grade 4

- Participants who develop a Grade 4 symptomatic toxicity will have study product permanently discontinued, and the site should notify the Principal Investigator within 72 hours.
- Participants experiencing Grade 4 toxicity requiring permanent discontinuation of study product should be followed up weekly until resolution of the AE or return to baseline. These participants will remain on study, off study treatment, and have all evaluations performed per the SoA.

Specific Management of Toxicities Related to Study-provided Drugs

HCQ

Gastrointestinal disturbance

Gastrointestinal disturbance (nausea, vomiting, dyspepsia, abdominal cramps, and diarrhea) is a common known possible side effect of HCQ. Taking with food or milk may improve tolerability.

Visual disturbances

Suspected visual changes should be evaluated for possible etiologies—if an HCQ-associated visual disturbance is suspected, HCQ should be stopped.

Allergic reactions

HCQ should be discontinued permanently if a serious allergic reaction is suspected. These participants will remain on study, off study treatment, and have all evaluations performed per the standard operating procedure(s).

QT prolongation

HCQ is associated with QT prolongation. QT will be assessed during the study using a portable monitor that can transmit electrocardiograms (ECGs) to measure the QTc to a central location, which will be monitored. Participants will be counseled about this risk.

Hypoglycemia

HCQ can be associated with hypoglycemia, particularly among patients with diabetes mellitus who are also taking insulin or sulfonylureas. Participants will be counseled about this risk.

Azithromycin

Gastrointestinal disturbance

Gastrointestinal adverse reactions (nausea, vomiting, abdominal pain, and diarrhea) are common side effect of azithromycin. Most gastrointestinal side effects do not require treatment discontinuation and participants may receive treatment for symptoms (e.g., antiemetics).

Allergic reaction

Serious allergic reactions, including, but not limited to anaphylaxis, angioedema, and dermatologic reactions such as Stevens-Johnson syndrome have been reported with azithromycin treatment. Azithromycin should be discontinued immediately in the event of a hypersensitivity reaction.

QT prolongation

Azithromycin is associated with QT prolongation. QT will be assessed during the study using a portable monitor that can transmit ECGs to measure the QTc to a central location, which will be monitored. Participants will be counseled about this risk.

LPV/r

QT prolongation

LPV/r may be associated with QT prolongation. Participants will be counseled about this risk.

Gastrointestinal disturbance

Nausea, vomiting, and diarrhea are common known possible side effects of LPV/r. Taking with food may improve tolerability. Participants will be counseled extensively about this risk. Ondansetron 4 mg tabs (#10) as needed for nausea and vomiting and loperamide 2 mg tabs (#10) as needed for diarrhea will be provided to each participant. A small supply will be given to participants for at least 2 days of symptom relief. It is anticipated that if symptoms are not controlled with the available medications, that participants may not be able to tolerate study medication. Loperamide is a well tolerated medication available over the counter. Risks include constipation, abdominal cramps. In rare cases, allergic reaction, fatigue/drowsiness, or toxic megacolon may occur. Up to 16 mg can be used per day. With higher than recommended doses, there is risk of QT prolongation. Participants will be instructed not to take more than 4 tablets (8 mg) of of loperamide per day. Those participants who take more than the recommended dosage to control diarrhea will have study medication held to reduce the risk of QT prolongation.

Ondansetron may be associated with headache, fatigue, and malaise. Constipation may also occur. Allergic reaction/hypersensitivity may also occur. Participants will be instructed not to take more than 12 mg per day of ondansetron. Those participants who take more than the recommended dose to control symptoms will have study medication held to reduce the risk of QT prolongation.

7.5. *Method of Treatment Assignment*

Participants will be randomized in a 1:1 ratio to placebo (ascorbic acid), or LPV/r, at the level of the household (all eligible participants in 1 household will receive the same intervention). The randomization plan will be overseen by the Study Statistician. The randomization code and resulting allocation list will be generated and overseen by the Study Statistician. The list will be blocked and stratified by site and risk level.

7.6. *Blinding*

7.6.1. *Patient Blinding*

This is a blinded study. However, the medications are not identical-appearing and dosing is different for the active agents so blinding will operate in different ways across those involved in the study, preserving blinding for the participants, those who directly interact with them, and measurement of the study endpoints:

- Participants will be blinded. The bottle of medication they receive will not identify the treatment allocation and only the number and frequency of pills to be taken. If >1 participant per household is randomized, all will receive the same treatment.
- Treating clinicians will be blinded, as the study medication will be dispensed directly to participants.
- Laboratory testing for viral shedding will be blinded, as laboratory staff will not be informed of randomized assignment. It should be noted that the viral shedding endpoint of the trial is an objective assessment, unlikely altered by unmasking, should it occur.
- Study pharmacy staff will be unblinded, as they will prepare the study medication.
- The study statistician will be unblinded for analysis purposes.

Eligible participants will receive one of the following therapies:

- Placebo: Ascorbic acid 1 gm orally twice on Day 1 followed by 500 mg orally twice daily for 9 days (Days 2 to 10)
- LPV/r 800 mg-200 mg orally twice on Day 1, followed by 400 mg-100 mg orally twice daily for an additional 9 days (Days 2 to 10)

HCQ and ascorbic acid will appear similar, and taste will be partially masked as HCQ can be bitter and ascorbic acid will be sour. However, LPV/r will not appear similar to ascorbic acid. Azithromycin and folic acid have the same dosing schedule but are different in shape. Because participants in 1 household will all receive the same assignment and study clinicians will not see the study medication, the blind will be maintained.

The participants will be blinded to their randomization group once assigned. At enrollment, the unblinded Study Pharmacist will use the randomization code revealed at the point of randomization to provide the participant with their group assignment and dispense the allocated study medication in a bottle marked with the study label. The medication and medication information, mid-nasal swabs sufficient to complete the study procedures, *DBS sampling kit, if within the sub-study,* and study instructions will be delivered to the participant.

7.7. *Preparation/Handling/Storage/Accountability*

Drugs should be stored at room temperature, as per package insert. Records must be maintained that document receipt, release for dosing, disposal, or return to the Sponsor.

7.8. *Treatment Compliance*

The participant will be contacted to ensure that they received the box of study supplies, were able to collect the first mid-nasal swab and store it appropriately, and took their first day of medication. Participants will be asked to complete a survey that includes information regarding treatment administration. Medication errors, including under-dosing, will be recorded in the survey. *In a sub-study, HCQ, azithromycin, and LPV/r concentrations via a DBS will also be evaluated.*

Consultation via Telehealth, text messaging, or telephone will be available to provide support to the participant to complete study procedures.

7.9. *Concomitant Therapy*

Participants will be asked about concomitant medications at the screening/baseline evaluation visit. During the study, participants will be asked to complete surveys (Daily Survey and Exit Contact Survey) that include information regarding any medication or vaccine (including over-the-counter or prescription medicines, vitamins, and/or herbal supplements) that the participant receives during the study. At each contact, the Investigator should question the participant about any medication taken.

7.9.1. *Prohibited Medications*

Medications listed as “Known Risk of Torsades de Pointe” on the website Credible Meds (CredibleMeds.org) will be prohibited with interventions known to prolong the QT interval (including HCQ, azithromycin, and LPV/r). These medications include: quinolone antibiotics (e.g., ciprofloxacin, levofloxacin, and moxifloxacin); macrolide antibiotics (e.g., erythromycin and clarithromycin); methadone; and antiarrhythmic drugs such as Class I (e.g., quinidine, procainamide, and disopromide), Class III (e.g., dofelitide, ibutilide, and sotalol), and amiodarone. Other prohibited medications include cisapride, cilastozol, cimetidine, fluconazole, terfenadine, antipsychotic medications (chlorpromazine, halperidol, and thioridazine), donepezil, oxaliplatin, and the antidepressants citalopram and escitalopram.

Prohibited medications for concomitant administration with HCQ include digoxin, cyclosporin, tamoxifen, anticoagulants (e.g., warfarin), some anticonvulsants, and rifampin. Refer to Appendix 5 for all medications prohibited for coadministration with HCQ.

Prohibited medications for concomitant administration with azithromycin include warfarin. Refer to Appendix 6 for all medications prohibited for coadministration with azithromycin.

Prohibited medications for concomitant administration with LPV/r include drugs that are highly dependent on CYP3A for clearance and for which elevated plasma concentrations are associated with serious and/or life-threatening reactions (e.g., alfuzosin, ranolazine, and simvastatin) and drugs that are potent CYP3A inducers where significantly reduced lopinavir plasma concentrations may be associated with the potential for loss of virologic response (e.g., apalutamide, rifampin, and St. John's Wort). Refer to Appendix 7 for all medications prohibited for coadministration with LPV/r.

7.9.2. *Precautionary Medications*

Use of medications classified as precautionary is not study prohibitory but will be discussed with the study clinician and the participant. These medications include androgens, antidiabetic agents, phenothiazines, artemether, beta-blockers, cardiac glycosides, aminoquinolines, dapsone, herbs, lumefantrine, maitake, mefloquine, monoamine oxidase inhibitors, nelfinavir, pegvisomant, prothionamide, and selective serotonin reuptake inhibitors. Careful monitoring is needed during concomitant use of azithromycin with colchicine or phenytoin. Medications listed as "Possible Risk of Torsades de Pointe" and "Conditional Risk of Torsades de Pointe" on the website Credible Meds (CredibleMeds.org) will be permitted as per discretion of study clinicians and after discussion with the study participant.

7.10. *Treatment After the End of the Study*

No additional treatment will be provided at the end of the study.

8. Discontinuation/Withdrawal Criteria

8.1. *Discontinuation of Study Treatment*

Study treatment will be discontinued for the following reasons:

- Hospitalization
- Requirement for prohibited concomitant medications or other contraindication to study product
- Occurrence of an AE requiring discontinuation of study product, including prolonged QT
- Request by participant to terminate study treatment
- Clinical reasons believed to be life-threatening by the physician, even if not addressed in Section 7.2

Participants who stop study product should continue study participation off study product with continued evaluations as per the SoA. The reason for study product discontinuation should be recorded.

Hospitalized participants will be followed through hospitalization and the last visit will be within 3 days of discharge.

8.2. *Withdrawal from the Study*

- A participant may withdraw from the study at any time at his/her own request or may be withdrawn at any time for the following reasons:
  - At the request of the primary care provider if he/she thinks the study is no longer in the best interest of the participant
  - Participant is judged by the Investigator to be at significant risk of failing to comply with the provisions of the protocol as to cause harm to self or seriously interfere with the validity of the study results
  - At the discretion of the Institutional Review Board/Ethics Committee or government agencies as part of their duties, Investigator, or industry supporter
- If the participant withdraws consent for disclosure of future information, the Sponsor may retain and continue to use any data collected before such withdrawal of consent.
- If a participant withdraws from the study, he/she may request destruction of any samples taken and not tested, and the Investigator must document this in the site study records.
- See SoA for data to be collected at the time of study discontinuation and follow-up and for any further evaluations that need to be completed.

8.3. *Lost to Follow-up*

A participant will be considered lost to follow-up if he/she is unable to be contacted by the study site.

The following actions must be taken if a participant fails to comply with required study procedures:

- The site must attempt to contact the participant as soon as possible and counsel the participant on the importance of maintaining the assigned procedure schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the Investigator or designee must 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.
- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

9. Study Encounters

The current COVID-19 pandemic has placed a significant burden on the healthcare system. For this study, specimen and data collection will be conducted to minimize impact of non-ill participants within the healthcare system. If the participant is assessed as eligible, contact between study participants and study personnel will occur via a Health Insurance Portability and Accountability Act (HIPAA)-compliant video conference (Telehealth). Optionally, contact with the study clinician or staff after Day 1 may be conducted by telephone.

Participants will be instructed to seek clinical care should they manifest any signs or symptoms of LRTI requiring medical intervention and notify their physician about trial participation.

9.1. *Screening/Baseline Evaluation: Day 0/1*

Participants will be assessed for study eligibility through a screening conducted through a web-based screening tool, HIPAA-compliant video conference (Telehealth), telephone, or text messaging.

Day 0 evaluations are as follows:

- Informed consent
- Collection of demographic information
- Sign HIPAA form
- Collection of past and current medical conditions, including known pregnancy and/or lactation status
- Collection of concomitant medication information
- Collection of information (including laboratory documentation of COVID-19 testing)
- Check of inclusion and exclusion criteria

Eligible participants will be randomized. Participants will receive a monitoring kit via courier, which will include thermometer and SpO2 device.They will be instructed on how to self-assess respiratory rate. In addition, they will receive a swab kit via courier or mail, which includes a study instruction booklet, swabs, plastic tubes for swab collection, a return box with affixed Category B UN3373 label, as required by International Air Transport Association (IATA) guidelines (UN3373 Medical Packaging 2020), and a pre-paid return shipping label.

The participant will do the following on Day 1:

- Collect mid-nasal swab for PCR
- Complete Daily Survey (online, telephone, or text messaging). This survey will include confirmation of adherence to study therapy dosing and daily swab collection, concomitant medication review, AEs, and symptoms review and objective measures including SpO2, respiratory rate, temperature, pulse, and ECG*. In addition, the modified inFLUenza patient-reported outcome (Flu-PRO) survey will be completed.
- Take study therapy (LPV/r or placebo [ascorbic acid]), as assigned
- *May collect DBS sample for analysis of HCQ, azithromycin, and LPV/r concentrations.*

Instructions for skin puncture and DBS sample preparation are provided in Appendix 9. A study team member will be available via Telehealth, telephone, or text messaging to provide support for completion of this study procedure.

Screening (Day 0) and Day 1 procedures can occur on the same day.

The SARS-CoV-2 positive test results will be confirmed through laboratory records or through detailed description of testing as described by participant.

9.2. *Day 2 Through Day 13*

The participant will do the following every day from Day 2 through Day 13, inclusive:

- Collect mid-nasal swab for PCR
- Take study therapy until Day 10 (inclusive) for LPV/r, or ascorbic acid (as assigned)
- Complete Daily Survey (online, telephone, or text messaging). This Survey will include confirmation of adherence to study therapy dosing and daily swab collection, concomitant medication review, AEs, and symptoms review and objective measures including SpO2, respiratory rate, temperature, pulse, and ECG. ECG measurements may be recorded on Days 2 through 11 only*. In addition, the modified Flu-PRO survey will be completed.
- *If in DBS sub-study, collect DBS samples for analysis of HCQ, azithromycin, or LPV/r concentrations at any time during this period (1 to 5 times) after study drug dosing has commenced.*
- **EKG monitoring may not be necessary if HCQ/AZ are not used.*

A study team member will be available via Telehealth, text messaging, or telephone to provide support for completion of study procedures. The courier will collect the swabs at minimum within 1 or 2 days of Day 7 and Day 13 and potentially as frequently as daily.

9.3. *Days 2 (±1 day), and Days 4, 9, and 14 (±2 days)*

Contact with study clinician or staff will be conducted via telemedicine (Telehealth) or telephone. Participants will be clinically assessed for signs and symptoms of respiratory distress and will have in person assessment of AEs. As needed, additional contact with the study clinician or staff will be conducted at the request of the participant (e.g., if developing concerning symptoms or an adverse event) or if needed to clarify study procedures or follow-up symptoms.

9.4. *Days 14 and 21*

The participant will do the following on Days 14 and 21:

- Collect mid-nasal swab for PCR
- Complete Daily Survey (online, telephone, or text messaging). This survey will include confirmation of adherence to study therapy dosing and daily swab collection, concomitant medication review, AEs, and symptoms review and objective measures including SpO2, temperature, and pulse. In addition, the modified Flu-PRO and WHO Ordinal Scale for Clinical Improvement surveys will be completed.

The courier will collect the swabs of Day 14 and Day 21 with the Day 28 swabs.

9.5. *Day 28*

The participant will do the following on Day 28:

- Collect mid-nasal swab for PCR
- Complete the Exit Contact Survey (online, telephone, or text messaging). This survey will include confirmation of adherence to study therapy dosing and daily swab collection, concomitant medication review, AEs, and symptoms review and objective measures including SpO2, temperature, and pulse. In addition, the modified Flu-PRO and WHO Ordinal Scale for Clinical Improvement surveys will be completed.
- *If in DBS sub-study, collect DBS sample for analysis of HCQ, azithromycin, or LPV/r concentrations as well as detection of anti-SARS-CoV-2 antibodies.*

For participants who are hospitalized on Day 28, the Day 28 procedures will be performed within 3 days after discharge from the hospital.

A study team member will be available via Telehealth, text messaging, or telephone to provide support for completion of study procedures.

The courier will collect the swabs at minimum within 1 or 2 days of Day 28.

Clinical outcomes will be confirmed through the electronic health record, if possible.

Participant Reimbursement

Participants will be reimbursed on Day 28 or at the post discharge visit. No reimbursement will be provided for unscheduled Telehealth visits requested by the participants for support with study procedures.

10. Study Assessments and Procedures

- Study procedures and their timing are summarized in the SoA.
- Protocol waivers or exemptions are not allowed.
- Immediate safety concerns should be discussed with the Sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study treatment.
- Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
- All baseline evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. The Investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.
- Procedures conducted as part of the participant's routine clinical management and obtained before signing of the informed consent form may be utilized for screening or baseline purposes provided that the procedures met the protocol-specified criteria and were performed within the time frame defined in the SoA.
- *Blood samples will only be collected as a part of a sub-study. The maximum amount of blood collected from each participant over the duration of the study, including any extra assessments that may be required, will not exceed 10 mL.*

10.1. *Efficacy Assessments*

10.1.1. *Mid-nasal Swab*

Participants will collect daily mid-nasal swabs for on Days 1 to 14, 21, and 28 for viral detection.

Participants will receive a swab kit either via courier or mail, which includes a study instruction booklet, swabs, plastic tubes for swab collection, a return box with affixed Category B UN3373 label, as required by IATA guidelines (UN3373 Medical Packaging 2020), and a pre-paid return shipping label. Participants are instructed to place their self-collected nasal swabs directly into the plastic tube that is pre-labeled with a unique barcode. Next, participants are instructed to place the plastic tube containing the self-collected nasal swab into a specimen bag, pre-packaged with an absorbent sheet, and then place the specimen bag into the provided return shipping box or pouch. Swab kits may contain either viral transport media, 0.9% saline, or phosphate-buffered saline, which have been shown to be equivalent for storage of SARS-CoV-2 samples based on validated assays (Rodino 2020). Previous testing has demonstrated that respiratory viral RNA is stable in room temperature for up to 1 week.

The used swabs will be collected by the courier or returned via the postal service or shipping service, such as FedEx.

Swabs will be subjected to RNA amplification and tested for SARS-CoV-2.

10.1.2. *Participant Survey*

Participants will be asked to complete surveys (Daily Surveys and Exit Contact Survey) that will include questions about symptoms from both the drug regimen and respiratory and systemic

symptom, review of concomitant medications, and other pertinent topics. A modified self-reported Flu-Pro instrument will be completed each day through Day 14 and on Day 21 and Day 28 to assess degree of illness (Yu 2020) (Appendix 12).

10.2. *Adverse Events*

Participants will be asked to complete surveys (Daily Surveys and Exit Contact Survey) that include information on any symptoms that they are experiencing. In addition, AE review by a staff member (via telephone, Telehealth, or text messaging) will be performed.

All AEs must be recorded on the electronic case report forms (eCRFs) if any of the following criteria have been met:

- All AEs meeting SAE definition
- All AEs leading to discontinuation of study medication
- All AEs judged by the site Investigator to be associated with study medication

10.2.1. *Adverse Events*

An AE is any untoward medical occurrence that occurs in a patient or clinical investigation subject administered a pharmaceutical product, which does not necessarily have to have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including abnormal laboratory findings), symptom, or disease temporally associated with the use of an investigational product, whether or not considered related to the product.

AEs may include the onset of new illness and the exacerbation of pre-existing medical conditions. An AE can include an undesirable medical condition occurring at any time, including baseline or washout periods, even if no study treatment has been administered.

10.2.2. *Serious Adverse Events*

An SAE is defined as any untoward medical occurrence that:

- Results in death
- Is life-threatening
- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is an important medical event that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require intervention to prevent one of the other outcomes listed in the definition above

All AEs that are recorded must have their severity graded. To grade AEs, sites should refer to the DAIDS AE Grading Table, corrected Version 2.1, July 2017, which can be found on the DAIDS Regulatory Support Center website at https://rsc.tech-res.com/docs/default-source/safety/division-of-aids-(daids)-table-for-grading-the-severity-of-adult-and-pediatric-adverse-events-corrected-v-2-1.pdf.

10.2.3. *Treatment-related AE and SAE*

A treatment-related adverse event is defined as any new AE that begins, or any preexisting condition that worsens in severity, after at least 1 dose of study treatment has been administered until the end of study (i.e,. Day 28) and is considered by the Investigator to be related to the study medication.

All AEs and SAEs should have attribution recorded as treatment- or not treatment-related, in the judgment of the site Investigator.

10.3. *Treatment of Overdose*

Overdose of HCQ, azithromycin, or LPV/r should be managed according to the labeling information (see Appendix 5, Appendix 6, and Appendix 7, respectively). Study drug overdose, including misuse or abuse of the product and medication errors, should be reported in the eCRF in the clinician notes.

Ascorbic acid and folic acid exhibit low toxicity; risks from overdose are expected to be minimal.

10.4. *Pregnancy*

Pregnancies occurring in participants enrolled in this study must be reported and followed to outcome. Pregnancy alone is not regarded as an AE unless there is a possibility that the study drug may have interfered with the effectiveness of a contraceptive medication. Elective abortions without complications should not be considered AEs unless they were therapeutic abortions. Hospitalization for normal delivery of a healthy newborn should not be considered an SAE. Pregnancy is not considered an SAE unless there is an associated serious outcome. Spontaneous abortions should always be reported as SAEs.

10.5. *Safety Assessments*

Safety will be assessed via participant Surveys, as shown in the SoA.

Participants will be asked to complete surveys (Daily Surveys and Exit Contact Survey) that include questions about their health, healthcare seeking, symptoms, illness within their household, contact, and mobility. Qualifying events will be recorded on the eCRF and reported as AEs, as described in Section 10.2.

10.6. *Dried Blood Spot Sub-study (Optional)*

All participants will be offered enrollment into the DBS sub-study. For those study sites not wishing to participate in the DBS sub-study, this will not be considered a protocol deviation.

Participants will receive instructions for DBS self-collection in writing, with telephone, Telehealth, and text messaging options as support. Once cards have been dried, they will be collected by the study courier and returned to the laboratory.

The aim of the DBS sub-study is to evaluate HCQ, azithromycin, and LPV/r drug concentrations as an adherence measure and the PK of HCQ, azithromycin, and LPV/r. If serological assays for SARS-CoV-2 are available, DBS may be tested for SARS-CoV-2 antibodies.

Pharmacokinetics

The exposure-response relationship of HCQ, azithromycin, or LPV/r of patients treated for SARS-CoV-2 has not been established. Population PK analyses can be used to further inform dose selection in other populations and support concentration-response investigations with efficacy and safety outcomes.

To accomplish this, sparse PK sampling techniques can be employed. This would involve collection of whole blood at 1 to 5 times after dosing has commenced. The time of collection post-dose can be random.

Given the long half-lives of HCQ and azithromycin, samples for several weeks after the last dose are also informative to the population PK model development.

The basic requirements for PK sampling are as follows:

1. Accurate record of time of the dose prior to the blood sampling (dd:mm:yy; hh:mm)
2. Accurate recording of time of blood sampling (dd:mm:yy; hh:mm) for each blood sampling
3. Whole blood can be obtained by venipuncture or capillary blood by skin puncture using a lancet
4. Approximately 100 µL of blood is then applied to filter paper as outlined in Appendix 9

Anti-SARS-CoV-2 Antibody Testing

DBS sample for serology will be collected at Day 28 and tested for SARS-CoV-2 antibodies provided that an appropriate test is available.

10.7. *Biohazard Containment*

As the transmission of SARS-CoV-2 and other respiratory droplet pathogens can occur through contact with respiratory droplets and contaminated surfaces, precautions will be employed by all personnel in the handling of all specimens for this study, as currently recommended by the Centers for Disease Control and Prevention and the National Institutes of Health.

All dangerous goods and materials, including diagnostic specimens and infectious substances, must be transported using packaging mandated by Code of Federal Regulations 42 Part 72. Please refer to instructions detailed in the IATA Dangerous Goods Regulations.

11. Statistical Considerations

11.1. *Sample Size Determination*

The overall size of the study is driven by the clinical endpoint, resolution of COVID-19 symptoms.

11.1.1. *COVID-19 Endpoints*

Overall sample size will depend on the number of events observed. To detect at least a 1.5-fold increased risk of the COVID-19 symptom resolution rate under a null hypothesis of no reduction

and four interim monitoring analyses with stopping for futility and efficacy based on the O'Brien-Fleming bounds and 80% power, the trial is designed to achieve 99 events per pairwise comparison for the symptom resolution endpoints. To achieve this, 173 participants will be enrolled per arm.

As the initial sample size target, 210 per arm was chosen for each experimental group (HCQ, HCQ + azithromycin), assuming a control arm rate of approximately 30% and 5% dropout rate.

HR (active/control)	Events/arm	N per arm	Median control arm (days)	Median active arm (days)*
1.4	142	247	10	7
1.5	99	173	10	7
1.6	75	129	10	6
1.7	59	103	10	6
1.8	49	85	10	6
1.9	41	71	10	5
2,0	36	63	10	5

*Under constant baseline hazard assumption

11.1.2. *Viral Shedding Endpoint*

Assuming the median time to cessation of viral shedding is 10 days and a constant baseline hazard for cessation of shedding, approximately 62% of participants would no longer experience viral shedding at 14 days under the null hypothesis of no intervention effect. Assuming 420 participants per pairwise comparison, 90% statistical power, 1-sided type I error rate of 2.5% for efficacy, we can detect at least a 37% increase in the rate of cessation of viral shedding. Under an extreme example of having only half that sample, 210, we could detect an increase of 54% or higher.

11.2. *Populations for Analyses*

For analysis purposes, the following populations are defined and will be described in greater detail in the trial Statistical Analysis Plan:

Population	Description
Intention to Treat (ITT)	All enrolled participants
Modified ITT	All enrolled participants who do not have LRTI at Baseline: high-risk group for the primary clinical endpoint; high- and low-risk groups (separately) for the primary virologic endpoint.
PK evaluable	Participants from the DBS sub-study with at least 1 interpretable PK sample.

DBS: dried blood spot; PK: pharmacokinetic; PCR: polymerase chain reaction; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2.

11.3. *Statistical Analyses*

The Statistical Analysis Plan will be developed and finalized before database lock and will describe the participant populations to be included in the analyses, the detailed analytical plans with endpoints and procedures for accounting for missing, unused, and spurious data. An Interim Monitoring Plan will also be developed to describe approaches for re-estimation of sample size and any planned interim analyses. This section presents a brief summary of the planned statistical analyses of the primary and secondary endpoints (these are listed in Section 4).

11.3.1. *Efficacy Analyses*

Demographic characteristics (age, sex, race) of each study group will be tabulated.

The mean age (plus range and standard deviation) by sex of the enrolled participants, as a whole and per group, will be calculated.

Primary analyses: The primary analyses will be conducted on the intend-to-treat population overall and by cohort (high-risk and low-risk). Participants randomized to each active treatment will be compared to participants randomized to placebo (ascorbic acid). If appropriate, the primary analysis will make use of all participants randomized to placebo, whether contemporaneously enrolled or not, using an Empirical Bayesian information borrowing method (described below). Due to anticipated heterogeneity in risk of disease progression, pre-specified baseline variables, including age, BMI, and days of symptoms at time of enrollment will be included in the model to increase precision.

Both primary endpoints will be analyzed using a Cox proportional hazards model with robust standard errors, adjusted for within household correlation.

As-treated analysis: The primary analyses will be repeated, replacing randomization arm by actual treatment to account for possible off-label use of and noncompliance to the investigational products.

Subgroup analyses: All subgroup analyses will be pre-specified in the Statistical Analysis Plan. Any further subgroup analyses will be considered ad hoc.

Missing Data

Due to the design of the study and retention activities, measurable outcomes are expected for all participants. However, in the unlikely event of a missing test result, the missing data will be imputed.

11.3.2. *Secondary Endpoints*

All secondary endpoints will be assessed in the Intention-to-Treat population overall and by cohort.

11.3.2.1. Safety Analyses

All safety analyses will be performed on the Intention-to-Treat population. AEs will be compared by study group.

11.3.2.2. Disease Severity

Disease severity, as measured by the WHO Ordinal Scale for Clinical Improvement (Appendix 11), will be compared between the groups using a proportional odds model.

Disease severity over the first 14 days and at Days 21 and 28 will also be measured by the modified Flu-PRO survey (Appendix 12). Results will be analyzed as for the WHO Ordinal Scale for Clinical Improvement.

Progression and regression of disease severity will be modeled using multistate models.

11.3.3. *Pharmacokinetic Analysis*

Sparse PK from DBS will be analyzed using standard population PK analysis methodologies using standard software such as NONMEM® version 7.4 or Phoenix NLME version 8.2.

11.3.4. *Exploratory Exposure-Response Analyses*

PK-evaluable participants will have post-hoc individual concentration profiles and exposure estimates determined for exploratory exposure-response analyses against primary and secondary efficacy and safety endpoints. Exploratory PK/pharmacodynamic analyses will be performed as the data allow.

11.3.5. *Combined Study Analysis*

This protocol is being published as a model protocol for other institutions to consider as they undertake studying treatments to prevent progression of COVID-19 among outpatients with SARS-CoV-2 infection. It is hoped that individual participant data from similar studies can be pooled into a combined study analysis. De-identified data from the present study will be made available for these purposes in accordance with the funder's open access policy (https://www.gatesfoundation.org/how-we-work/general-information/open-access-policy).

12. References

Ballow CH, Amsden GW. Azithromycin: the first azalide antibiotic. Ann Pharmacother. 1992;26(10):1253-61.

Barrett B, Brown R, Rakel D, et al. Placebo effects and the common cold: a randomized controlled trial. Ann Fam Med. 2011 Jul-Aug;9(4):312-322.

Belouzard S, Millet JK, Licitra BN, et al. Mechanisms of coronavirus cell entry mediated by the viral spike protein. Viruses. 2012 Jun;4(6):1011-33.

Cai J, Xu J, Lin D, et al. A Case Series of children with 2019 novel coronavirus infection: clinical and epidemiological features. Clin Infect Dis. 2020 Feb. pii: ciaa198.

Cao B, Wang Y, Wen D, et al. A trial of lopinavir–ritonavir in adults hospitalized with severe Covid-19. N Engl J Med. 2020 Mar 18. doi: 10.1056/NEJMoa2001282 [Epub ahead of print].

Chan KS, Lai ST, Chu CM, et al. Treatment of severe acute respiratory syndrome with lopinavir/ritonavir: a multicenter retrospective matched cohort study. Hong Kong Med J. 2003;9:399-406.

Chen F, Chan KH, Jiang Y, et al. In vitro susceptibility of 10 clinical isolates of SARS coronavirus to selected antiviral compounds. J Clin Virol. 2004;31:69-75.

Chen J. Pathogenicity and transmissibility of 2019-nCoV-a quick overview and comparison with other emerging viruses. Microbes Infect. 2020 Mar;22(2);69-71.

Chen N, Zhou M, Dong X, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. Lancet. 2020 Feb;395(10223):507-513.

Chorin E, Matthew D, Shulman E et al. The QT interval in patients with SARS-CoV-2 infection treated with hydroxychloroquine/azithromycin. 2020. preprint doi: https://doi.org/10.1101/2020.04.02.20047050.

Chu CM, Cheng VC, Hung IF, et al. Role of lopinavir/ritonavir in the treatment of SARS: initial virological and clinical findings. Thorax. 2004;59:252-256.

Collins K, Jackson K, Gustafson D. Hydroxychloroquine: a physiologically based pharmacokinetic model in the context of cancer-related autophagy modulation. J Pharmacol Exp Ther. 2018;365(3):447-459.

Fujii T, Luethi N, Young PJ, et al. Effect of vitamin C, hydrocortisone, and thiamine vs hydrocortisone alone on time alive and free of vasopressor support among patients with septic shock: the VITAMINS randomized clinical trial. JAMA. 2020 Jan 17. doi: 10.1001/jama.2019.22176. [Epub ahead of print].

Furst DE, Lindsley H, Baethge B, et al. Dose-loading with hydroxychloroquine improves the rate of response in early, active rheumatoid arthritis: a randomized, double-blind six-week trial with eighteen-week extension. Arthritis Rheum. 1999 Feb;42(2):357-365.

Gao J, Tian Z, Yang X. Breakthrough: chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. Biosci Trends. 2020 Feb. doi: 10.5582/bst.2020.01047.

Gautret P, Lagier JC, Parola P, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. Int J Antimicrobial Agents. 2020; Epub 2020 Mar.

Graat J, Schouten E, Kok F. Effect of daily vitamin E and multivitamin-mineral supplementation on acute respiratory tract infections in elderly persons: a randomized controlled trial. JAMA. 2002 Aug;288(6):715-721.

Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. 2020 Mar; Cell. pii: S0092-8674(20)30229-4 (pre-publication).

Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet. 2020 Feb;395(10223):497-506.

Keyaerts E, Vijgen L, Maes P, et al. In vitro inhibition of severe acute respiratory syndrome coronavirus by chloroquine. Biochem Biophys Res Commun. 2004 Oct;323(1):264-268.

Lee JS, Adhikari NKJ, Kwon HY, et al. Anti-Ebola therapy for patients with Ebola virus disease: a systematic review. BMC Infect Dis. 2019 May;19(1):376.

Li CC, Wang XJ, Wang HR. Repurposing host-based therapeutics to control coronavirus and influenza virus. Drug Discov Today. 2019 Mar;24(3):726-736.

Linton NM, Kobayashi T, Yang Y, et al. Incubation period and other epidemiological characteristics of 2019 novel coronavirus infections with right truncation: a statistical analysis of publicly available case data. J Clin Med. 2020 Feb;9(2). pii: E538.

Mackenzie AH. An appraisal of chloroquine. Arthritis Rheum. 1970 May-Jun;13(3):280-291.

Menzel M, Akbarshahi H, Bjermer L, et al. Azithromycin induces anti-viral effects in cultured bronchial epithelial cells from COPD patients. Sci Rep. 2016 Jun;6:28698.

Poschet JF, Perkett EA, Timmins GS, et al. Azithromycin and ciprofloxacin have a chloroquine-like effect on respiratory epithelial cells. 2020. bioRxiv preprint. doi: https://doi.org/10.1101/2020.03.29.008631.

Rodino KG, Espy MJ, Buckwalter SP, et al. Evaluation of saline, phosphate buffered saline and minimum essential medium as potential 2 alternatives to viral transport media for SARS-CoV-2 testing. J Clin Microbiol. 2020. 2020 Mar 30.

Rolain JM, Colson P, Raoult D. Recycling of chloroquine and its hydroxyl analogue to face bacterial, fungal and viral infections in the 21st century. Int J Antimicrob Agents. 2007 Oct;30(4):297-308.

Savarino A, Shytaj IL. Chloroquine and beyond: exploring anti-rheumatic drugs to reduce immune hyperactivation in HIV/AIDS. Retrovirology. 2015 Jun;12:51.

Schoenfeld D, Zheng H, Finkelstein D. Bayesian design using adult data to augment pediatric trials. Clin Trials. 2009;6:297-304.

Schrezenmeier E, Dörner T. Mechanisms of action of hydroxychloroquine and chloroquine: implications for rheumatology. Nat Rev Rheumatol. 2020 Mar;16(3):155-166.

Shippey EA, Wagler VD, Collamer AN. Hydroxychloroquine: an old drug with new relevance. Cleve Clin J Med. 2018 Jun;85(6):459-467.

Slater AF. Chloroquine: mechanism of drug action and resistance in Plasmodium falciparum. Pharmacol Ther. 1993 Feb-Mar;57(2-3):203-235.

Srinivasa A, Tosounidou S, Gordon C. Increased incidence of gastrointestinal side effects in patients taking hydroxychloroquine: a brand-related issue? J Rheumatol. 2017 Mar;44(3):398.

Tett SE, Cutler DJ, Day RO, Brown KF. Bioavailability of hydroxychloroquine tablets in healthy volunteers. Br J Clin Pharmacol. 1989;27(6): 771-779.

Touret F, Gilles M, Barral K, NougairédeA, Decroly E, de Lamballerie X, Coutard B. *In vitro* screening of a FDA approved chemical library reveals potential inhibitors of SARS-CoV-2 replication. 2020 bioRxiv preprint doi:https://doi.org/10.1101/2020.04.03.023846.

UN3373 Medical Packaging. Biological Substance Category B. Available at: https://www.un3373.com/category-biological-substances/category-b. Updated 2018 Jun. Accessed 2020 Mar 01.

Vincent MJ, Bergferon E, Benjannet S, et al. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. Virol J. 2005 Aug;2:69-78.

Wang M, Cao R, Zhang L, et al. Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. Cell Res. 2020 Mar;30(3):269-271.

White House Press Release. https://www.whitehouse.gov/presidential-actions/proclamation-declaring-national-emergency-concerning-novel-coronavirus-disease-covid-19-outbreak. 2020 Mar 13.

WHO News Release. Shortage of personal protective equipment endangering health workers worldwide. 2020 Mar 03.

WHO News Release. Statement on the second meeting of the International Health Regulations (2005) Emergency Committee regarding the outbreak of novel coronavirus (2019-nCoV). 2020 Jan 30.

Wrapp D, Wand N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. Science. 2020 Mar 13;367(6483):1260-1263.

Yao X, Ye F, Zhang M, et al. In vitro antiviral activity and projection of optimized dosing design of hydroxychloroquine for the treatment of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Clin Inf Dis. 2020 Mar. pii: ciaa237.

Yu J, Powers JH, Vallo D, et al. Evaluation of efficacy endpoints for a Phase IIb study of a respiratory syncytial virus vaccine in older adults using patient-reported outcomes with laboratory confirmation. Value Health. 2020 Feb;23(2):227-235.

Zhu N, Zhang D, Wang W, et al. A novel coronavirus from patients with pneumonia in China, 2019. N Engl J Med. 2020 Feb;382(8):727-733.

Wu C-Y, Jan J-T, Ma S-H, et al. Small molecules targeting severe acute respiratory syndrome human coronavirus. Proc Natl Acad Sci U S A. 2004;101 10012-10017.

WHO. Report of the WHO-China joint mission on coronavirus disease 2019 (COVID-19). 2020 Feb 16-24.

13. Appendices

Appendix 1: Abbreviations and Terms

Term	Definition
ACE2	Angiotensin converting enzyme 2
AE	Adverse event
CER	Control event rate
COVID-19	Coronavirus disease
CQ	Chloroquine
CYP	Cytochrome P450
CYP3A	Cytochrome P450 3A
DAIDS	Division of Acquired Immunodeficiency Syndrome
DBS	Dried blood spot
DSMB	Data and safety monitoring board
EC_{50}	Half-maximal effective concentration
ECG	Electrocardiogram
eCRFs	Electronic case report forms
Eligible	Qualified for enrollment into the study based upon adherence to inclusion/exclusion criteria
FDA	Food and Drug Administration
Flu-PRO	InFLUenza patient-reported outcome
GCP	Good Clinical Practice
GFR	Glomerular filtration rate
HCQ	Hydroxychloroquine
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human immunodeficiency virus
IATA	International Air Transport Association
ICF	Informed consent form
Index case	Term used throughout the protocol to denote the person with confirmed or suspected SARS-CoV-2 infection to whom the study participant was exposed
IRB	Institutional Review Board
LPV/r	Lopinavir-ritonavir
LRTI	Lower respiratory tract infection
MERS-CoV	Middle East respiratory syndrome coronavirus
Participant(s)	Term used throughout the protocol to denote the enrolled individual(s)
PBPK	Physiologically based pharmacokinetics
PCR	Polymerase chain reaction

PEP	Post-exposure prophylaxis
PK	Pharmacokinetic(s)
QTc	Heart-rate corrected QT interval
RNA	Ribonucleic acid
SAE	Serious adverse event
SARS-CoV	Severe acute respiratory syndrome coronavirus
SARS-CoV-2	Severe acute respiratory syndrome coronavirus 2
SoA	Schedule of Activities
SpO2	Oxygen saturation
USPI	United States prescribing information
WHO	World Health Organization
WIRB	Western Institutional Review Board

Appendix 2: Protocol Structure

The protocol for this trial will be structured to be modular. Given the perpetual nature of this trial, there may be modifications and/or additions to the main protocol. Each modification and/or addition to the main protocol will be subject to the data and safety monitoring board who will receive recommendation from other study committees (see Appendix 3).

The main protocol will contain all the background and rationale for this trial and all generic information to the trial, the research approach, the trial design and conduct, and the overall trial governance, and ethical considerations. Other appendices for study governance, site-specific protocol addendum template, hydroxychloroquine (HCQ) label, azithromycin label, lopinavir-ritonavir label, pharmacokinetic modelling, and pharmacokinetic sample collection and analysis are provided below. The Intervention-Specific Appendices (e.g., HCQ label appendix) will contain the information about the interventions. There will be a specific appendix for each of the intervention arm with features of the given intervention strategy and how it will be delivered, and any additional endpoints and data collection that are not covered in the Main Protocol.

Appendix 3: Study Governance Considerations

Investigators and Institutional Affiliations

The following Investigators and Institutional Affiliations were established at the time of protocol authoring. Designees may be provided, as appropriate. Other institutions may utilize this model protocol with permission from the Principal Investigator.

Name	Role	Institution	Contact Information
Dr. Christine Johnston, MD, MPH	Principal Investigator	University of Washington	Phone: +1 206 520 4318 Email: cjohnsto@uw.edu
University of Washington Coordinating Center			
Dr. Elizabeth R. Brown, ScD	Study Statistician	Fred Hutchinson Cancer Research Center	Phone: +1 206 667-1731 Email: erbrown@fredhutch.org
Dr. Jared Baeten, MD, PhD	Co-Investigator	University of Washington	Phone: +1 206 520-3808 Email: jbaeten@uw.edu
Dr. Connie Celum, MD, MPH	Co-Investigator	University of Washington	Phone: +1 206 520-3800 Email: ccelum@uw.edu
Dr. Ruanne Barnabas, MD, PhD	Co-investigator	University of Washington	Phone: +1 206 520 3813 Email: rbarnaba@uw.edu
Dr. Joshua Schiffer, MD	Co-investigator	Fred Hutchinson Cancer Research Center	Phone: +1 206 667 7359 Email: jschiffe@fredhutch.org

Committees Structure

The Trial Steering Committee will take the overall responsibility for the trial design and conduct. All committees will act in accordance to the International Clinical Harmonization Guidelines for Good Clinical Practice (GCP) Principles.

<u>Study Team Monitoring</u>

The study team will monitor the conduct of the study through monthly summary reports of arms of accrual and baseline characteristics and quarterly reports of data pooled over treatment arms of data completeness, specimen collection, and adverse events (AEs). The study team will review individual participant-level safety data frequently to assess the relation of all reported AEs to study treatment. On a weekly basis, the study team will review by-arm summaries of premature study discontinuations and premature study treatment discontinuations (and reasons) and AEs.

Independent Monitor

Study conduct will be monitored by an independent monitor. Monitors will visit participating clinical research sites, or schedule equivalent virtual visits, to review the individual participant records, including consent forms, electronic case report forms, supporting data, laboratory specimen records, and endpoints through laboratory and medical records (physicians' progress notes, nurses' notes, individuals' hospital charts), to ensure protection of study participants, compliance with the protocol, and accuracy and completeness of records. The monitors also will inspect the sites' regulatory files to ensure that regulatory requirements are being followed and the sites' pharmacies to review product storage and management.

Data and Safety Monitoring Board

An independent data and safety monitoring board (DSMB) will be convened for this study with expertise in coronavirus disease (COVID-19) or respiratory viruses, antiviral therapies and shedding, and emerging epidemics and a biostatistician. The purpose of the DSMB is to monitor the study for operational futility, social harms, and efficacy. The DSMB will evaluate the progress of the project, including periodic assessments of accrual, retention, safety, performance and variation of the project sites, and other factors that can affect project implementation.

The DSMB will review and approve modifications to the overall enrollment target based on the event rate. Due to the anticipated rapid speed of enrollment and the short duration of the study, it is unlikely that pre-specified stopping rules for efficacy and futility in terms of the efficacy of hydroxychloroquine (HCQ), HCQ + azithromycin, or lopinavir-ritonavir will be reached before all participants are enrolled. The DSMB will review severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) endpoints.

The DSMB will conduct interim reviews (if specified) when adequate data have been accrued and convene by teleconference. Open reports containing accrual and retention rates, participant characteristics, and serious adverse events will be sent to the protocol team and DSMB members the week prior to the DSMB meeting. Only the DSMB members and the unblinded biostatistician will receive password-protected closed reports of SARS-CoV-2 endpoints by randomization arm.

Regulatory and Ethical Considerations

The study will be conducted according to GCP, the Belmont Report, and the Declaration of Helsinki. The study protocol, site-specific informed consent forms (ICFs), participant education and recruitment materials, and other requested documents—including any subsequent modifications—will be reviewed and approved by Western Institutional Review Board (WIRB), as the single IRB of record, responsible for oversight of research conducted at the study sites. Subsequent to initial review and approval, the WIRB will review the study at least annually.

Informed Consent Process

The principles of informed consent in the current edition of the Declaration of Helsinki will be implemented in each clinical study before any protocol-specified procedures or interventions are carried out. The consent form will describe the purpose of the study, the procedures to be followed, and the risks and benefits of participation. A copy of the consent form will be given to the participant, and this fact will be documented in the participant's record.

A participant who is rescreened is not required to sign another ICF; eligibility for the study must be re-checked prior to enrollment.

Study Records

Each study site will establish a standard operating procedure for confidentiality protection. Each site will ensure that study records including administrative documentation and regulatory documentation as well as documentation related to each participant enrolled in the study, including ICFs, locator forms, case report forms, notations of all contacts with the participant, and all other source documents are stored in a secure manner.

Confidentiality

Participants' study information will not be released without their written permission, except as necessary for oversight by:

- The protocol Principal Investigators or designees
- Study funders
- WIRB
- University of Washington IRB

All laboratory specimens, evaluation forms, reports, and other records that leave the site will be identified by coded number only to maintain participant confidentiality. The exceptions are SARS-CoV-2 testing results, which are subject to local and state reporting which is name-based. Local public health may contact participants diagnosed with SARS-CoV-2 for the purpose of surveillance and contact notification. Participants will be informed prior to SARS-CoV-2 testing that results are reportable and may lead to contact by local public health if results are positive for infection.

All records will be kept locked. After initial acquisition of data, all computer entry and networking programs will be done with coded numbers only. Clinical information will not be released without written permission of the participant, except as necessary for monitoring by the IRB, Office for Human Research Protections, other local, United States, and international regulatory entities as part of their duties or the industry supporters or designees.

Appendix 4: Site-Specific Protocol Addendum Template

PURPOSE: The use of this site-specific protocol addendum is recommended with a multi-site study.

The purpose of this site-specific protocol addendum is to obtain information describing the local site-specific elements for conduct of the Core Protocol. The descriptions should focus on who, what, and where of the local study activities.

INSTRUCTIONS:

1. Add the Name of the study site to the Title of the Protocol in the header of the document.
2. Please complete each section below. If general description language already appears in the Core Protocol, which adequately describes the local activity at the study site, a notation can be inserted in the given section that reads, "As described in the Core Protocol."
3. Please identify the completed addendum document using a version number and date.
4. Please note: the site-specific protocol addendum will require review and approval by the local Institutional Review Board (IRB)/Ethics Committee (EC) of record for the study site.

Study Site Information:

Name of Institution/Company:

Address:

Study Site Information for the Site Investigator:

Name of Site Investigator:

Title:

Institution/Affiliation:

Address:

Telephone Number:

Cell/Other Number:

Fax Number:

Study Site Number:

Assurance Information:

United States Department of Health and Human Services Office for Human Research Protections Assurance Federal Wide Assurance number:

Expiration Date:

IRB/Ethics Committee Information:

Name of Human Subjects Protection oversight office for study site:

Name and number of reviewing IRB/EC:

Telephone Number at Office:

Fax Number at Office:

If Available, Point of Contact at IRB/EC:

Name:

Telephone Number:

Local Site-specific Information:

1. Identify key study personnel (include name, title, address, point of contact information).
2. Describe the key study personnel roles and responsibilities.
3. Describe the local recruiting procedures and strategies. Provide copy of any site-specific recruitment material. Identification of personnel responsible for completing tasks.
4. Describe the local consenting process. Provide a copy of the site-specific consent form(s). If an Ombudsman is named for the study site, provide name, title, and point of contact information.
5. Identify local study collaborations at the site such as pharmacy, laboratories, and other institutional departments.
6. Describe the local specimen/sampling procedures in place. Include acquisition, disposition, storage, and unique coding. If samples will be kept for future use, describe procedures and the security measures for short-term and long-term management. Provide name of repository.
7. Describe the plan for on-site management of study records and data, and participant study records. Explain procedures and security measures in place for short- and long-term management. Declare who will have access to data.
8. Describe the local measures in place to promote privacy and confidentiality.
9. Describe the local procedures in place for provision of care for the participant regarding research-related injuries.
10. Identify who the participant can contact locally should the participant have any questions regarding the research. Identify who the participant can contact should the participant have questions regarding their rights as a study participant. Include the points of contact information (name, title, and telephone number).
11. Describe the procedures in place to address Health Insurance Portability and Accountability Act requirements. If a separate authorization form will be used at the

study site, provide a copy or ensure appropriate language has been included in the consent document(s).

12. Describe any unique site-specific study procedures or supplemental activities.
13. Declare any unique study population/cultural influences, socioeconomic conditions, etc.
14. Declare any other site-specific reporting obligations and procedures. Name any additional oversight boards or committees.
15. Define abbreviations that may apply to the specific study site.

Appendix 5: Hydroxychloroquine Label

Generic hydroxychloroquine label (current as of June 2018) is available online at https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=b82bbda6-64f2-4426-b4ec-254eeea895ae and provided below.

Appendix 6: Azithromycin Label

The Azithromycin label is available online at http://labeling.pfizer.com/ShowLabeling.aspx?format=PDF&id=511 and provided below.

Appendix 7: Lopinavir-ritonavir Label

Kaletra (Lopinavir-ritonavir), US Prescribing Information, AbbVie, December 2019 is available online at https://rsc.niaid.nih.gov/sites/default/files/Lopinavir-Ritonanir%20%28Kaletrata%29%20PI%20dated%20December%202019.pdf and provided below.

Appendix 8: Physiologically Based Pharmacokinetic Model for Azithromycin Dose Regimen

A physiologically based pharmacokinetic (PBPK) model for azithromycin was developed previously and verified against clinical data (Johnson et al., 2016). With the exception of the B:P ratio, which was set at 2.28, the input parameters for the azithromycin model remained the same. This updated model was then used to predict exposures in plasma and lung after administration of 500 mg for 1 day and 250 mg QD for 4 days, a dosage regimen that was used by Gautret et al. (2020) to prevent bacterial super-infection in patients receiving hydroxyl-chloroquine as treatment for SARS-CoV-2. Based on physicochemical data, the predicted lung to tissue ratio (Kp) was 16.33, which was lower than the observed ratio of 50.5 (Lucchi et al., 2008). Both predicted and observed ratios were used in simulations (100 subjects aged 20 – 50 years; 50% female) of plasma and lung concentration-time profiles which were then compared against *in vitro* values of 2.12 (EC_{50}) and 8.65 µM (EC_{90}) reported for inhibition of SARS-CoV-2 by azithromycin (Touret et al., 2020) (Figure 1). It should be noted that the unbound fraction in plasma (fu) was set at 0.69 and the predicted fu in lung tissue was 0.82. Thus, unbound and total concentrations were similar in both plasma and lung.

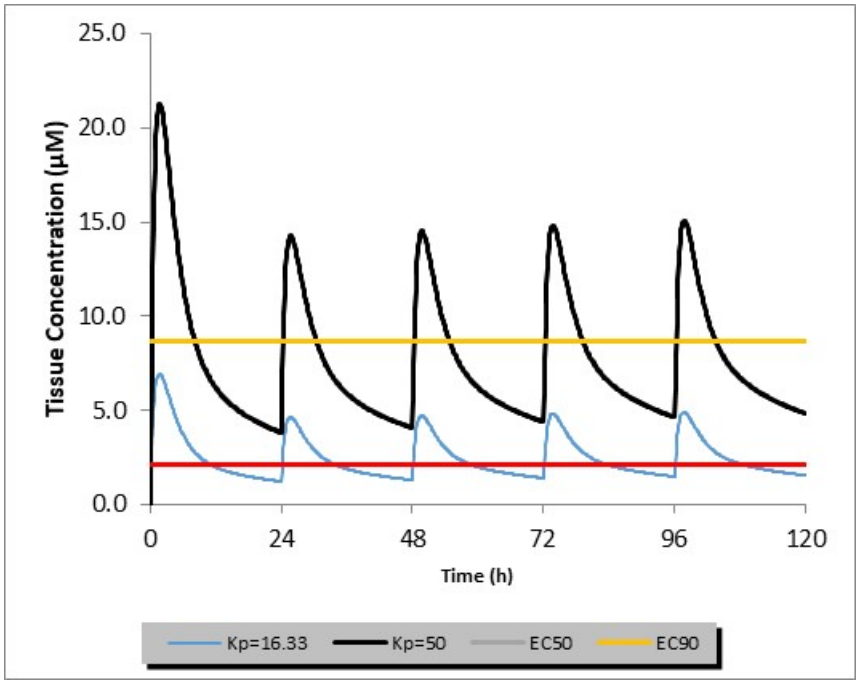

Figure 1. Predicted concentration-time profiles for azithromycin in the lung based on Kp values of 16.33 and 50 in the lung.

Based on the simulations shown, it appears that the dosing regimen of 500 mg for 1 day followed by 250 mg QD for 4 days, is able to attain concentrations of azithromycin in lung that would approach the *in vitro* EC_{50} value determined for azithromycin.

Gautret et al. (2020) Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomised clinical trial. International Journal of Antimicrobial Agents – In Press 17 March 2020: – DOI : 10.1016/j.ijantimicag.2020.10594.

Johnson TN, Jamei M and Rowland-Yeo K (2016). How does in vivo biliary elimination of drugs change with age? Evidence from in vitro and clinical data using a systems pharmacology approach. *Drug Metab Dispos*; 44:1090-1098.

Lucchi M, Damle B, Fang A et al., (2008). Pharmacokinetics of azithromycin in serum, bronchial washings, alveolar macrophages and lung tissue following a single oral dose of extended or immediate release formulations of azithromycin. Journal of Antimicrobial Chemotherapy; 61: 884–891.

Touret F, Gilles M, Barral K, NougairédeA, Decroly E, de Lamballerie X, Coutard B. *In vitro* screening of a FDA approved chemical library reveals potential inhibitors of SARS-CoV-2 replication. bioRxiv preprint doi: https://doi.org/10.1101/2020.04.03.023846.

Appendix 9: Pharmacokinetic Sample Collection and Analysis

Collection of Blood

Skin puncture

1. Put on a pair of disposable gloves.
2. Before skin puncture, the participant should warm his/her hands. The finger is massaged anterogradely to enrich the blood flow toward the puncture site.
3. Clean the skin of the palmar side of the tip of the distal phalanx of the third or fourth finger of the nonwriting hand with a suitable disinfectant, for example, 70% isopropyl alcohol. Puncture the skin by a single-use safety lancet. The finger should be held in such a position that the gravity facilitates the collection of blood from the fingertip.
4. When collection of capillary blood by skin puncture is complete, place a bandage on the fingertip.

Venipuncture

To be conducted by trained personnel per standard procedures.

Preparation of Blood Spots

Preparation from blood collected by skin puncture

1. Wipe off the first drop of blood with a gauze pad because it may contain excess tissue fluids. Massage the finger again to increase blood flow at the puncture site. Transfer the following drop to one of the circles of a filter card without touching the surface directly with the fingertip. Allow the blood to be soaked into the texture of the filter by capillary forces only.
2. Let the next large drop of capillary blood form on the fingertip and collect it in the next circle. Continue this procedure until all necessary circles are filled or blood flow stops.
3. Do not squeeze or "milk" the finger excessively if the blood flow is not sufficient to fill all the required circles of the filter card. If blood flow stops, place a bandage on the fingertip. Perform a second skin puncture on another finger if more blood is needed for the examination.
4. For blood obtained by venipuncture, use syringe to apply approximately 100 µL of blood on the filter paper.

Drying of Blood Spots

To dry the blood spots, put the filter cards on a clean paper towel and let them dry, preferably overnight (but for at least 4 hours), at room temperature in the absence of any external source of heat. When the drying process is complete, the blood spots have a uniformly dark brownish color and no red areas are visible anymore.

Storage and Transportation of Dried Blood Spots

NOTE: Processing of the blood spots can be interrupted after drying. The filter cards can now be stored.

1. For storage, put the filter paper card in a single, gas-impermeable zipper bag, containing 1 to 2 desiccant sachets to protect the specimens from moisture. Optionally, add a humidity indicator card.
2. Transfer this bag to a freezer as soon as possible. If freezers are not available under field conditions, storage at -4°C or even at ambient temperature is feasible for up to 14 days.
3. Transport frozen dried blood spots specimens on dry ice. For filter cards initially kept at ambient temperature, use a triple packaging system, which consists of the zipper bag(s) as the inner container(s) as well as an inner and an outer envelope. No content markings are required on the outer envelope for shipment by regular mail, but the international biohazard symbol must be affixed to the primary inner container.
4. Exclude the filter cards from further processing if the desiccant packs and/or the additional humidity indicator card changes to a pink color.

On the filter paper, the participant should record the following:

1. Date of collection

Samples that appear to be collected according to the schedule of activities, which have the required amount of blood for a 5- to 6-mm punch and have the minimal required information (1 through 3 above), will be processed for hydroxychloroquine (and metabolite) and azithromycin concentration.

Appendix 10: Physiologically Based Pharmacokinetic Modeling of Hydroxychloroquine Used for Post-exposure Prophylaxis

Appendix 11: WHO Ordinal Scale for Clinical Improvement

WHO COVID-19 Core Protocol
SOLIDARITY Trial
Version 10.0, March 22, 2020

Patient State	Descriptor	Score
Uninfected	No clinical or virological evidence of infection	0
Ambulatory	No limitation of activities	1
	Limitation of activities	2
Hospitalized Mild disease	Hospitalized, no oxygen therapy	3
	Oxygen by mask or nasal prongs	4
Hospitalized Severe Disease	Non-invasive ventilation or high-flow oxygen	5
	Intubation and mechanical ventilation	6
	Ventilation + additional organ support – pressors, RRT, ECMO	7
Dead	Death	8

Appendix 12: The inFLUenza Patient-Reported Outcome instrument (Flu-PRO) – Modified for SARS-CoV-2

DAILY SURVEY (D1-14, D21, D28)

inFLUenza patient-reported outcome (Flu-PRO): 32 items validated, can score across domains.

***FYI: This takes about 5 minutes to complete

How are you feeling today?

FLU-PRO	Not at all	A little bit	Somewhat	Quite a bit	Very much
Nose					
Runny or dripping					
Congestion or stuffy					
Sneezing					
Sinus pressure					
Lack of smell					
Throat					
Sore throat					
Scratchy or itchy throat					
Difficulty swallowing					
Lack of taste					
Eyes					
Teary or watery eyes					
Sore or painful eyes					
Eyes sensitive to light					
Chest/Respiratory					
Trouble breathing					
Chest congestion					
Chest tightness					
Dry or hacking cough					
Wet or loose cough					

Sputum (coughing up sputum or phlegm)

Wheezing

Gastrointestinal

Felt nauseous

Stomach ache

Vomit

Diarrhea

Body/Systemic

Felt Dizzy

Head congestion

Headache

Lack of appetite

Sleeping more than usual

Body aches or pains

Weak or tired

Chills of shivering

Felt cold

Felt hot

Sweating

How are you feeling today?

0 no symptoms 1 Mild 2 Moderate 3 Severe 4 Very severe

Please rate interference in daily activities due to illness:

1 Not at all 2 A little bit 3 Somewhat 4 Quite a bit 5 Very much

How is your general health?

1 Poor 2 Fair 3 Good 4 Very good 5 Excellent

Have you returned to your usual health today? Yes/no

Have you returned to your usual activities today? Yes/no

TAKE MEDS, TAKE SWAB (Copy post-exposure prophylaxis)

Take your vitals:

- Oxygen level
- Pulse
- Temperature
- Respiratory rate
- Electrocardiogram monitor

PM Time:

- Take meds
- Take vitals
- Oxygen level
- Pulse
- Temperature
- Respiratory rate

Anxious mood	Not at all	mild	moderate	severe	very severe

Yu J, Powers JH, Vallo D, et al. Evaluation of efficacy endpoints for a Phase IIb study of a respiratory syncytial virus vaccine in older adults using patient-reported outcomes with laboratory confirmation. Value Health. 2020 Feb;23(2):227-235.