Open-label, randomized clinical trial of hydroxychloroquine alone or hydroxychloroquine plus azithromycin or chloroquine alone or chloroquine plus azithromycin in the treatment of SARS CoV-2 infection

Version Number: 2.0
March 30 2020

STATEMENT OF COMPLIANCE

The study will be carried out in accordance with Good Clinical Practice (GCP) as required by the following:

- U.S. Code of Federal Regulations applicable to clinical studies (45 CFR 46)
- 21 CFR 312
- ICH GCP E6
- Completion of Human Subjects Protection Training

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments, and provides the necessary assurances that this clinical study will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH guidelines.

Signed: _____ Date_____

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Open-label, randomized clinical trial of hydroxychloroquine alone or hydroxychloroquine plus azithromycin or chloroquine alone or chloroquine plus azithromycin in the treatment of SARS CoV-2 infection

PROTOCOL SUMMARY

Subjects: Non-ventilated hospitalized participants with SARS-CoV-2 infection

Study Site: Washington University in St Louis/ Barnes Jewish Hospital

Study Duration: 2 years

Number of subjects: 500

Subject Participation Duration: 6 weeks

Description of Study Design: Open-label block randomized study with four arms:

Arm 1. Hydroxychloroquine alone
Arm 2. Hydroxychloroquine *plus* azithromycin
Arm 3. Chloroquine alone
Arm 4. Chloroquine *plus* azithromycin

Primary

1. Time (hours) from randomization to recovery defined as 1) absence of fever, as defined as at least 48 hours since last temperature ≥ 38.0°C without the use of fever-reducing medications AND 2) absence of symptoms of greater than mild severity for 24 hours AND 3) not requiring supplemental oxygen beyond pre-COVID baseline AND 4) freedom from mechanical ventilation or death

Secondary

1. Time to resolution of fever defined as at least 48 hours since last temperature ≥ 38.0°C without the use of fever-reducing medications
2. Time to improvement in symptoms (scored as mild or absent and remained so for 24 hour)
3. Mean improvement in symptom from baseline
4. Duration of hospitalization
5. Proportion requiring supplementary oxygen (above baseline usage) at any time during follow-up
6. Duration (days) of requirement for supplementary oxygen
7. Proportion requiring ICU admission at any time during follow-up
8. Proportion requiring mechanical ventilation at any time during follow-up
9. Ventilator free days
10. All-cause mortality

BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

Coronavirus disease 19 (COVID-19) is a rapidly emerging pandemic caused by severe acute respiratory syndrome coronavirus 2 (SARS CoV-2). At the time of writing, over half a million people have tested positive for the virus, approximately one sixth of those are located in the US with the numbers steadily rising. At present the overall case fatality rate is estimated between 2-7% with significant variation reported between settings[1]. The most common symptoms are fever, a non-productive cough, malaise and shortness of breath[2]. The rapid emergence of the syndrome globally in combination with a population that does not appear to have any previous exposures has led to a rapid utilization of healthcare resources. In many hardest hit locations this has led to rationing of healthcare, and a severe strain of healthcare workers as they struggle to provide care to a significantly increased patient caseload.[3]

At present there are no approved drugs for the management of SARS CoV-2 although a number of trials worldwide are actively recruiting. Several candidate drugs have been identified through data derived from the SARS CoV outbreak in the early 2000s as potential candidates for treatment of SARS-CoV-2 and have limited preliminary data for their use in SARS CoV-2 infection. Both Chloroquine and hydroxychloroquine have *in vitro* activity against SARS-CoV-2[4] and have been used extensively in the past for management of other medical conditions including autoimmune disorders and malaria prophylaxis and treatment. However, despite the limited data on the clinical benefit of these agents many international and regional guidelines are recommending their use[1]. Recent limited reports from China showed that chloroquine can shorten symptoms, as well as reduce viral load[5,6] In a small French study, a combination of hydroxychloroquine and azithromycin reportedly appeared to be superior to hydroxychloroquine alone however the data was generated from a very small trial[6].

Based on pre-clinical evidence of effectiveness and evidence of safety from long-time clinical use for other indications, we propose to perform a phase III trial of hospitalized non-ventilated participants to assess if hydroxychloroquine and chloroquine in combination with azithromycin vs each drug alone improves the time to symptom resolution.

STUDY AIMS/ENDPOINTS

Primary

1. Time (hours) from randomization to recovery defined as 1) absence of fever, as defined as at least 48 hours since last temperature ≥ 38.0°C without the use of fever-reducing medications AND 2) absence of symptoms of greater than mild severity for 24 hours AND 3) not requiring supplemental oxygen beyond pre-COVID baseline AND 4) freedom from mechanical ventilation or death

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5. Proportion requiring supplementary oxygen (above baseline usage) at any time during follow-up
6. Duration (days) of requirement for supplementary oxygen
7. Proportion requiring ICU admission at any time during follow-up
8. Proportion requiring mechanical ventilation at any time during follow-up
9. Ventilator free days
10. All-cause mortality

Exploratory

1. COVID-19 specific mortality
2. Number of patients with diagnosed cardiomyopathy
3. Number of patients with new onset arrhythmia

STUDY DESIGN

This Phase III trial will utilize four treatment strategies in non-critically ill participants (not requiring ICU admission and/or mechanical ventilation) with SARS CoV-2 infection, Participants will receive hydroxychloroquine or chloroquine with or without azithromycin. The primary outcome will be time from randomization to recovery

Enrollment

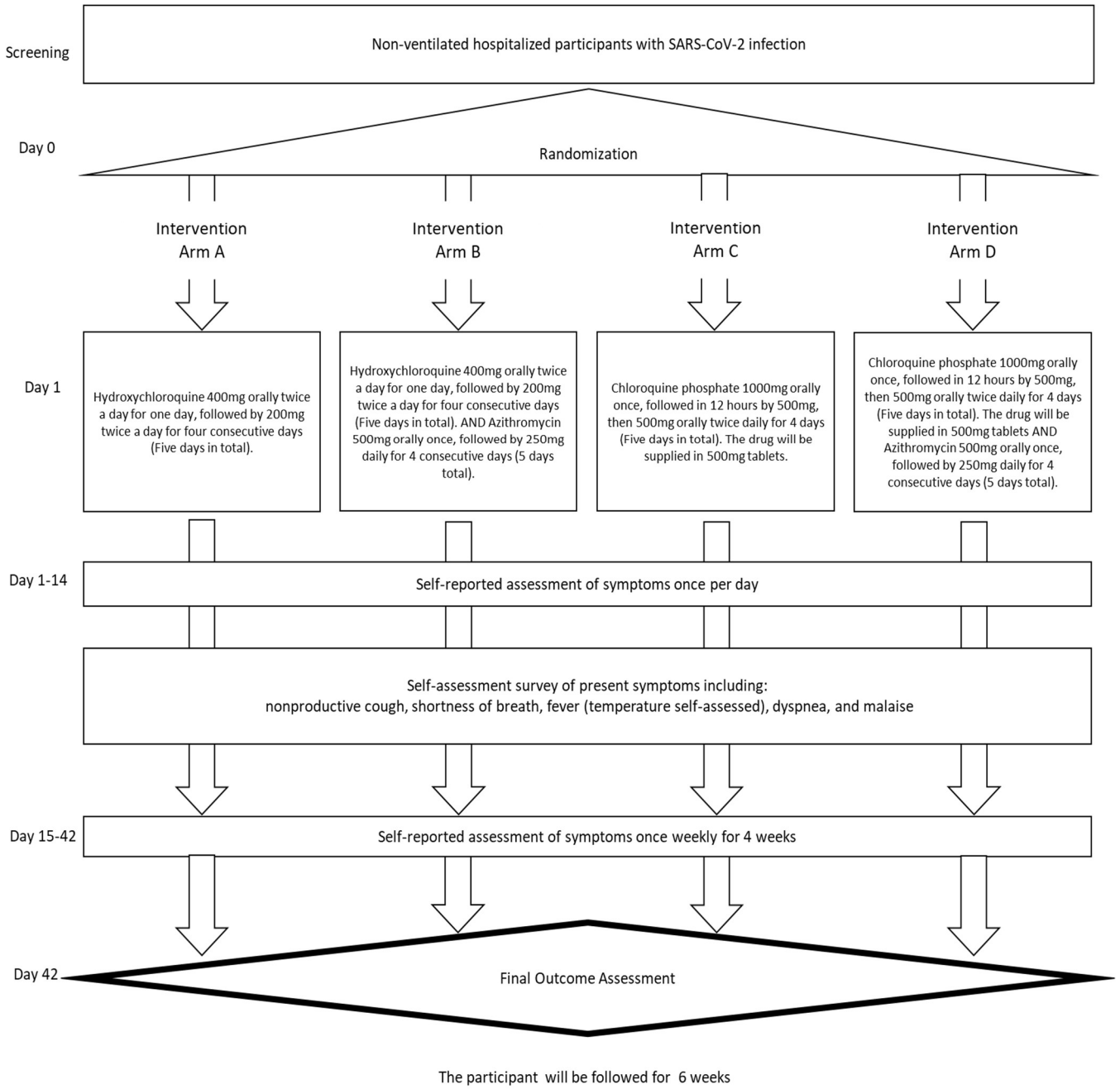
Hospitalized patients with documented SARS CoV-2 infection will be screened for study eligibility. If eligible, consenting participants will be randomized to one of the four arms. Up to 125 participants will be recruited for each of the study groups, with a planned total enrollment of 500 participants. Participants will be randomized to either one of the following arms in a 1:1:1:1 manner. Drugs will be provided by the BJC investigational pharmacy using standardized Epic order sets. If participants are discharged, we will coordinate with the investigational pharmacy services to ensure that participants receive the full course of therapy.

Arm 1: Hydroxychloroquine 400mg orally twice a day for one day, followed by 200mg twice a day for four consecutive days (Five days in total). The drug will be supplied in 200mg tablets.

Arm 2: Hydroxychloroquine 400mg orally twice a day for one day, followed by 200mg twice a day for four consecutive days (Five days in total). The drug will be supplied in 200mg tablets.
AND
Azithromycin 500mg orally once, followed by 250mg daily for four consecutive days (five days total). The drug will be supplied in 250mg tablets.

Arm 3: Chloroquine phosphate 1000mg orally once, followed in 12 hours by 500mg, then 500mg orally twice daily for 4 days (Five days in total). The drug will be supplied in 500mg tablets.

Arm 4: Chloroquine phosphate 1000mg orally once, followed in 12 hours by 500mg, then 500mg orally twice daily for 4 days (Five days in total). The drug will be supplied in 500mg tablets.
AND
Azithromycin 500mg orally once, followed by 250mg daily for 4 consecutive days (5 days total). The drug will be supplied in 250mg tablets.



SUBJECT INCLUSION CRITERIA

- Hospitalization for management of SARS CoV-2 infection
- Positive SARS CoV-2 test
- Age >=18 years
- Provision of informed consent
- Electrocardiogram (ECG) ≤48 hours prior to enrollment
- Complete blood count, glucose-6 phosphate-dehydrogenase (G6PD), comprehensive metabolic panel and magnesium ≤48 hours prior to enrollment from standard of care.
- If participating in sexual activity that could lead to pregnancy, individuals of reproductive potential who can become pregnant must agree to use contraception throughout the study. At least one of the following must be used throughout the study:
    - Condom (male or female) with or without spermicide
    - Diaphragm or cervical cap with spermicide
    - Intrauterine device (IUD)
    - Hormone-based contraceptive

SUBJECT EXCLUSION CRITERIA

- Contraindication or allergy to chloroquine, hydroxychloroquine or azithromycin
- Current use hydroxychloroquine, chloroquine or azithromycin
- Concurrent use of another investigational agent
- Invasive mechanical ventilation
- Participants who have any severe and/or uncontrolled medical conditions such as:
    - unstable angina pectoris,
    - symptomatic congestive heart failure,
    - myocardial infarction,
    - cardiac arrhythmias or know prolonged QTc >470 males, >480 female on ECG
    - pulmonary insufficiency,
    - epilepsy (interaction with chloroquine),
- Prior retinal eye disease
- Concurrent malignancy requiring chemotherapy
- Known Chronic Kidney disease, eGFR<30 or dialysis
- G-6-PD deficiency, if unknown requires G6PD testing prior to enrollment
- Known Porphyria
- Known myasthenia gravis
- Currently pregnant or planning on getting pregnant while on study
- Breast feeding
- AST/ALT >five times the upper limit of normal ULN*
- Bilirubin >five times the ULN*
- Magnesium <1.4 mEq/L*
- Calcium <8.4mg/dL >10.6mg/dL*
- Potassium <3.5 >5.5 mEg/L*
- Current concomitant use of contraindicated drugs including antiarrhythmics, antidepressant, anticonvulsants (see Appendix 2 for full list)

*Patients who do not meet laboratory criteria may be rescreened within 48 hrs.

Discontinuation criteria

Discontinuation of treatment

- Lack of efficacy, defined as deterioration of the clinical condition or delayed response requiring, in the opinion of the investigator, alternative therapy
- Occurrence of an AE that, in the opinion of the investigator, warrants discontinuation of the subject from the study drug
- Pregnancy

Discontinuation of study participation

- Withdrawal of consent;
- Investigator or Sponsor decision that withdrawal is in the subject's best interest
- Lost to follow up (every attempt should be made to contact the subject). Three attempts will be made to contact participants electronically or by phone. If unsuccessful, a letter will be sent to the participant to encourage follow up. Thereafter if a response is not received in 14 days the participant will be considered lost to follow up.

POTENTIAL RISKS AND BENEFITS

Potential risks:

Drug associated risks

Risks of chloroquine:

The following adverse reactions have been identified during post-approval use of Chloroquine or other 4-aminoqunoline compounds. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to reliably estimate their frequency or establish a causal relationship to drug exposure.

Ocular disorders: Maculopathy and macular degeneration have been reported and may be irreversible. Irreversible retinopathy with retinal pigmentation changes (bull's eye appearance) and visual field defects (paracentral scotomas) in patients receiving long-term or high-dosage 4-aminoquinoline therapy have been reported. Visual disturbances (blurring of vision and difficulty of focusing or accommodation); nyctalopia; scotomatous vision with field defects of paracentral, pericentral ring types, and typically temporal scotomas (e.g., difficulty in reading with words tending to disappear, seeing half an object, misty vision, and fog before the eyes) have been reported. Reversible corneal opacities have also been reported.
Immune system disorders: Urticaria, anaphylactic reaction including angioedema.
Ear and labyrinth disorders: Nerve type deafness; tinnitus, reduced hearing in patients with preexisting auditory damage.
Musculoskeletal and connective tissue-disorders: Sensorimotor disorders, skeletal muscle myopathy or neuromyopathy leading to progressive weakness and atrophy of proximal muscle groups, depression of tendon reflexes and abnormal nerve conduction.
Gastrointestinal disorders: Hepatitis increased liver enzymes, anorexia, nausea, vomiting, diarrhea, abdominal cramps.
Skin and subcutaneous tissue disorders: Erythema multiforme, Stevens-Johnson syndrome, toxic epidermal necrolysis, exfoliative dermatitis. Pleomorphic skin eruptions, skin and mucosal

pigmentary changes; lichen planus-like eruptions, pruritus,; drug rash with eosinophilia and systemic symptoms (DRESS syndrome); photosensitivity and hair loss and bleaching of hair pigment.
Blood and lymphatic system disorders: Pancytopenia, aplastic anemia, reversible agranulocytosis, thrombocytopenia and neutropenia. Hemolytic anemia in G6PD deficient patients.
Nervous system disorders: Convulsions, mild and transient headache, polyneuropathy, acute extrapyramidal disorders (such as dystonia, dyskinesia, tongue protrusion, torticollis).
Neuropsychiatric disorders: Neuropsychiatric changes including psychosis, delirium, anxiety, agitation, insomnia, confusion, hallucinations, personality changes, depression, and suicidal behavior.
Cardiac disorders: Hypotension, electrocardiographic changes (particularly, inversion or depression of the T-wave with widening of the QRS complex), and cardiomyopathy (which may result in cardiac failure and in some cases a fatal outcome).
Cardiac arrhythmias, conduction disorders such as bundle branch block / atrio-ventricular block, QT interval prolongation, torsade de pointes, ventricular tachycardia and ventricular fibrillation have been reported with therapeutic doses of Chloroquine as well as with overdose. The risk is greater if Chloroquine is administered at high doses. Fatal cases have been reported.
Metabolic and Nutritional disorders: Hypoglycemia.
Risk of overdose: Chloroquine is very rapidly and completely absorbed after ingestion. Toxic doses of Chloroquine can be fatal. As little as 1 g may be fatal in children. Toxic symptoms can occur within minutes. The symptoms of overdosage may include nausea, vomiting, headache, drowsiness, visual disturbances, cardiovascular collapse, convulsions, hypokalemia, rhythm and conduction disorders including QT prolongation, torsades de pointes, ventricular tachycardia and ventricular fibrillation, followed by sudden potentially fatal respiratory and cardiac arrest. Immediate medical attention is required, as these effects may appear shortly after the overdose. Cases of extrapyramidal disorders have also been reported in the context of Chloroquine overdose.

Risks of hydroxychloroquine
Rare (1% to 10%):
Vision loss due to retinopathy which may be irreversible

Frequency not defined:
Central nervous system: Ataxia, dizziness, emotional disturbance, emotional lability, headache, irritability, lassitude, nervousness, nightmares, psychosis, seizure, sensorineural hearing loss, suicidal tendencies, vertigo
Dermatologic: Acute generalized exanthematous pustulosis, alopecia, bleaching of hair, bullous rash, dyschromia (skin and mucosal; black-blue color), erythema multiforme, erythema of skin (annulare centrifugum), exacerbation of psoriasis (nonlight sensitive), exfoliative dermatitis, lichenoid eruption, maculopapular rash, morbilliform rash, pruritus, skin photosensitivity, Stevens-Johnson syndrome, toxic epidermal necrolysis, urticaria
Endocrine & metabolic: Exacerbation of porphyria, weight loss
Gastrointestinal: Anorexia, diarrhea, nausea, stomach cramps, vomiting
Hematologic & oncologic: Agranulocytosis, anemia, aplastic anemia, hemolysis (in patients with glucose-6-phosphate deficiency), leukopenia, purpuric rash, thrombocytopenia
Hepatic: Acute hepatic failure (rare), hepatic insufficiency (rare)
Hypersensitivity: Angioedema

Neuromuscular & skeletal: Myopathy (including palsy or neuromyopathy, leading to progressive weakness and atrophy of proximal muscle groups; may be associated with mild sensory changes, loss of deep tendon reflexes, and abnormal nerve conduction)
Ophthalmic: Accommodation disturbance, corneal changes (transient edema, punctate to lineal opacities, decreased sensitivity, deposits, visual disturbances, blurred vision, photophobia [reversible on discontinuation]), decreased visual acuity, macular edema, nystagmus disorder, optic disc disorder (pallor/atrophy), retinal pigment changes, retinal vascular disease (attenuation of arterioles), retinitis pigmentosa, scotoma, vision color changes, visual field defect
Otic: Tinnitus
Respiratory: Bronchospasm

<1%, postmarketing, and/or case reports: Cardiomyopathy, epithelial keratopathy (Dosso 2007), hypoglycemia (Cansu 2008; Unübol 2011), macular degeneration, maculopathy

Risks of azithromycin:
Common (>10%):
Gastrointestinal: Loose stools (≤14%; single-dose regimens tend to be associated with increased incidence), vomiting (children, single-dose regimens tend to be associated with increased incidence: 1% to 14%; adults: ≤2%; adults, single 2 g dose: 1% to 7%), diarrhea (2% to 9%; single-dose regimens 4% to 14%), nausea (≤7%; high single-dose regimens 4% to 18%)

Rare (1% to 10%):
Cardiovascular: Chest pain (≤1%), palpitations (≤1%)
Central nervous system: Dizziness (≤1%), drowsiness (≤1%), fatigue (≤1%), headache (≤1%), vertigo (≤1%)
Dermatologic: Skin rash (≤5%; single-dose regimens tend to be associated with increased incidence), dermatitis (children: ≤2%), pruritus (≤2%), skin photosensitivity (≤1%)
Endocrine & metabolic: Increased lactate dehydrogenase (1% to 3%), increased gamma-glutamyl transferase (1% to 2%), increased serum potassium (1% to 2%), decreased serum bicarbonate (adults: ≥1%), decreased serum glucose (adults: >1%)
Gastrointestinal: Abdominal pain (1% to 7%; single-dose regimens tend to be associated with increased incidence), anorexia (≤2%), dysgeusia (≤1%), dyspepsia (≤1%), flatulence (≤1%), gastritis (≤1%), melena (adults, multiple-dose regimens: ≤1%), mucositis (≤1%), oral candidiasis (≤1%)
Genitourinary: Vaginitis (≤3%), genital candidiasis (adults, multiple-dose regimens: ≤1%)
Hematologic & oncologic: Decrease in absolute neutrophil count (children: 15% to 16%; 500 to 1,500 cells/mm^3), decreased hematocrit (adults: >1%), decreased hemoglobin (adults: >1%), increased neutrophils (adults: >1%), thrombocythemia (adults: >1%), change in neutrophil count (children: ≥1%), eosinophilia (≥1%), lymphocytopenia (≥1%)
Hepatic: Increased serum ALT (≤6%), increased serum AST (≤6%), increased serum bilirubin (≤3%), cholestatic jaundice (≤1%)
Neuromuscular & skeletal: Increased creatine phosphokinase (1% to 2%)
Renal: Increased serum creatinine (≤6%), increased blood urea nitrogen (≤1%), nephritis (adults, multiple-dose regimens: ≤1%)
Respiratory: Bronchospasm (≤1%)
Miscellaneous: Fever (children: (≤2%)

Participants will be fully consented and aware of the potential risks associated with their participation in the study and have the knowledge that they can discontinue the study at any

time. Before a patient is approached, permission will be received from the treating physician. Consent will be performed by a physician co-investigator or be delegated to a coordinator under the supervision of a physician co-investigator. A physician co-investigator will review and approve all enrollments. Consents may be performed utilizing electronic informed consent as possible, but paper informed consent will be available as well. Each subject will be monitored carefully and appropriately and will be instructed to contact the study team promptly upon noticing any complication or health issues. Should this occur, subjects' health history will be reviewed by the study team.

All subjects asked to participate in the clinical studies will be advised that their participation in the studies is entirely voluntary, that they can choose discontinue participation at any time and that this will not negatively impact on their access to routine health care.

For participants enrolled under a legal authorized representative (LAR), the PI for the project, or a designated research coordinator, will maintain the enrolled subject's name and date of birth in a secured electronic format. Throughout the patient's stay, they will periodically review if the participant has recovered the ability to understand the nature of study participation and to make his/her own medical decisions. When this occurs, the enrolled subject will be provided a signed copy of the consent document that was originally signed by and provided to the LAR, informed that they were enrolled in the study, and the section regarding withdrawal of consent will be specifically pointed out to them if they choose to do that. Furthermore, they will be allowed to ask questions regarding their participation in the study and the purpose of the study from the research coordinator or PI during that encounter. For participants that are scheduled to be placed in a LTAC facility or any other facility who are still unable to consent prior to transfer, the PI or designated research coordinator will make every effort to have a second copy of the signed consent form provided to the LAR and to inform the LAR once again that the participant or the LAR can contact the PI in the future at any time as described in the consent form to discuss the study or to withdraw consent.

Other risks to staff and research participants
Given the known contagious potential for COVID-19, we will take several precautions to limit risks to staff and other participants following federal, state, local and university guidelines. We will ensure that we are following WU/HRPO guidelines as they are updated.

Known Potential Benefits

Preliminary data suggests that hydroxychloroquine or chloroquine used alone or in combination with azithromycin could improve outcomes in SARS CoV-2 infection.

RANDOMIZATION PROCEDURES
Randomization will be performed by the BJH Investigational Drug Service (IDS) using a computer-generated randomization sequence, which will be concealed from investigators and the research team through study completion. Consented patients will be randomly assigned to one of four open-label treatment cohorts in a 1:1:1:1 ratio using permutation blocks (block size will be determined by the study statistician). Medications will be prepared and dispensed by the IDS pharmacy and delivered to the appropriate coordinator. The research pharmacy will maintain randomization, dispensing, and accountability logs. All medications will be stored in a

secure location within the research pharmacy and maintained under controlled environmental conditions.

Handling of Withdrawals

Subjects are free to withdraw from the study at any time.

Subjects may be taken off study without their consent if the study doctor determines that it is in the subject's best interest not to continue to participate in the study. In addition, the study doctor may remove a subject from study participation if the subject is unable to complete the required study procedures, or if the study is stopped by the institution, the sponsor, or the Food and Drug Administration (FDA) or other health authorities. If the subject is removed from the study, the Principal Investigator or designee will contact the participant to discuss the study stopping procedures.

Participants who withdraw are withdrawn or terminated from the study, or are lost to follow-up after study enrollment visit will not be replaced.

STUDY PROCEDURES

Schedule of events

	Screen (using SOC in last 48hr	Baseline Screening and baseline may be combined if labs are done							Weeks			
Week		1	1	1	1	1	1	1-2	3	4	5	6
Day		0	1	2	3	4	5	6-14	21	28	35	42
Window (hrs) +/-				6	6	6	6	24	48	48	48	48
Informed consent	x											
SARS CoV-2	x											
EKG	x											
Medication review	x											
Complete blood count, comprehensive metabolic panel and magnesium	x											
History of Glucose-6 phosphate-dehydrogenase	x											

(G6PD) deficiency assessment												
(G6PD) lab (¥research if preformed)	x¥											
Determine eligibility		x										
Pregnancy test Unless SOC within 7-14 days	x											
Study drug		x	x	x	x	x	x					
Review medical record**	x	x	x	x	x	x	x	x	x	x	x	x
Adverse Drug Reaction Collection			x	x	x	x	x	x	x	x	x	x
Symptom screen		x	x	x	x	x	x	x	x	x	x	x
Temperature (from medical record taken and recorded by participant while at home*		x	x*	x*	x*	x*	x*	x*				

* thermometer at home and log

** medical history, study outcomes, AEs, SAEs

Pregancy Test

Unless done as SOC within the last 7-14 days prior to entry a negative β-HCG (human chorionic gonadotropin) pregnancy test (urine or serum) on day of enrollment for women presumed to be of reproductive will be done.

Eligibility

Participants hospitalized with SARS CoV-2 infection who meet the inclusion/exclusion criterion will be enrolled. Standard of care investigations including laboratory test (complete blood count, comprehensive metabolic panel, magnesium, and glucose-6-phosphate dehydrogenase) and ECG performed within 48 hours will be used to determine study eligibly.

Baseline assessment

Eligible participants will be consented and randomized according to the procedure outlined above. Demographics and clinical parameters will be recorded

Follow up

Participants will be asked to complete daily symptom surveys for 14 days from randomization, then weekly thereafter for 4 weeks resulting in a total duration of follow up of 42 days. During hospitalization, daily symptom surveys will be completed in conjunction with the study coordinators via telephone. On discharge participants will have the option to complete electronic symptom surveys or complete symptom surveys via telephone with study coordinator. In the event that the participant opts for electronic symptom surveys on discharge they will also

receive a follow up call from a study coordinator every 7 days during the initial 14 day period. In addition, failure to submit a symptom survey will prompt a study follow up call.

On discharge from hospital in addition to completing daily symptom surveys participants will be asked to take and record their temperature twice daily with extra recordings performed if concern for fever onset.

Symptoms will be collected using the Flu-PRO scale (appendix 1) validated for use in influenza[7] as at present validated tools for COVID-19 are not available.

To assure safety and protocol adherence phone calls to participants will be initiated if there is no contact at the designated time points.

End of study visit
The final study visit will via telephone at 6 weeks post enrollment

Study Duration

Total duration of participation in the study should not exceed 6 weeks

SAFETY ASSESSMENTS

ADVERSE EVENTS AND STUDY MONITORING
Definition of Adverse Events
Adverse Event (AE)

An Adverse Event is any untoward medical occurrence observed in a patient that develops or worsens from baseline status in association with a subject's participation in the research, whether considered research-related or not. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, disease, or injury temporally associated with the research, whether or not considered related to the research.

Serious Adverse Event (SAE)

A Serious Adverse Event is any AE that results in any of the following outcomes:

- Death
- A congenital anomaly, birth defect, or cancer in a baby born to a female subject
- Life-threatening adverse experience (places the subject at immediate risk of death from the event as it occurred)
- Inpatient hospitalization or prolongation of existing hospitalization
- Persistent or significant disability/incapacity
- Based on appropriate medical judgment, may jeopardize the subject's health and may require medical or surgical intervention to prevent one of the other outcomes listed in this definition

Adverse Drug Effect (ADE)

An Adverse Drug Effect is an AE that is definitely, probably, or possibly related to the use of study drug

Unexpected Adverse Drug Effect (UADE)

An Unexpected Adverse Drug Effect is any serious adverse effect on health or safety, or any life- threatening problem or death caused by, or associated with study drug, if that effect, problem, or death was not previously identified in nature, severity, or degree of incidence in the investigational plan or application; or any other unanticipated serious problem associated with a drug that relates to the rights, safety, or welfare of study participants.

Outcome

The clinical course of all AEs should be followed until a medical outcome is determined. The clinical outcome of all AEs will be recorded as follows:

- Fatal
- Not Yet Recovered (Patient did not recover and symptoms/sequelae continue)
- Recovered (Patient returned to baseline status)
- Recovered with Sequelae (Patient recovered but with clinical sequelae from the event)
- Unknown

Adverse Event Collection Requirements for this protocol

All AEs must be recorded on the eCRFs if any of the following criteria have been met.

- All Grade ≥2 AEs
- All AEs that led to a change in study treatment regardless of grade
- All AEs meeting SAE definition
- Started within 7 days after the last dose of study drug

All AEs that are reported must have their severity graded. To grade AEs, refer to the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events (DAIDS AE Grading Table), corrected Version 2.1, July 2017.

Serious Adverse Events (SAEs)

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 a congenital anomaly/birth defect

- Is an important medical event that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed in the definition above).

Reporting Requirements

Adverse events must be reported by study personnel into the Redcap system using the electronic case report forms within 3 days of their knowledge of the occurrence. Investigators are responsible for following all relevant local IRB policies and procedures. The Principal Investigator will follow IRB policy in terms of reporting adverse events.

Serious Adverse Events that occur within 6 weeks after randomization must be reported by the site investigator or personnel into the Redcap system on the Serious Adverse Event CRF within 1 working day after the site investigator or personnel first learn of the event.
For SAEs, the Principal Investigator will review the report and obtain any needed additional clarifications concerning the event by direct telephone conversations and/or e-mail with the involved providers. The investigator's description and categorization of the event, and additional relevant information, will then be reviewed by the study's Safety Officer. The Safety Officer will determine if the event was properly categorized. If not, he will provide the Principal Investigator with a modified categorization, along with a brief written rationale for re-categorizing the event. In nearly all instances, the Safety Officer's categorization will be considered the final categorization of the event.
Expedited Reporting

Events meeting criteria as an unanticipated problem (UP) or unexpected adverse drug event (UADE) will be reported to the IRB within 10 working days of becoming aware of the event
If the event results in death, it will be reported within 1 working day of becoming aware of the event
All other events that do not meet criteria as a UP or UADE will be reported to WUSTL IRB at the time of continuing review

Independent Data and Safety Monitoring Board (IDSMB)
An IDSMB consisting of members who are independent from the Sponsor will be established. The IDSMB will have at least 3 members. The IDSMB will be responsible for reviewing outcomes and AEs. The first IDSMB meeting will be held after approximately 100 participants have completed treatment. The frequency of subsequent meetings will be decided by the IDSMB, but will be not less than every 3 months. The IDSMB may also meet in *ad hoc* meetings at its discretion as needed in response to events occurring in the trial. Additional responsibilities for the Committee include confirmation of the safety and appropriateness of the selected dosing regimen. After each meeting, the IDSMB will recommend that the study can continue, or may recommend changes to the study or stopping the study. Details of the membership and responsibilities of the IDSMB will be included in the IDSMB Charter.

The IDSMB will also monitor new developments in COVID-19 research and treatment, and factor any related ethical considerations into its determinations as to whether or not the study should be terminated for futility for one.

STATISTICAL CONSIDERATIONS

The proposed study is a 4-arm parallel group, open-label randomized clinical trial. The treatment arms, hydroxychloroquine alone (HCQ), hydroxychloroquine + azithromycin (HCQ + AZM), chloroquine alone (CQ) and chloroquine + azithromycin (CQ + AZM) will be randomly assigned eligible participants. The primary hypotheses in this study are whether the addition of AZM to either HCQ or CQ improve patient recovery rates.

Primary endpoint analysis
The primary endpoint of this study is the time to recovery following the definition outlined in the STUDY AIMS/ENDPOINTS section. Our primary analysis will be to use Kaplan-Meier (KM) survival analysis to test the statistical null hypothesis that there is no difference in recovery rates between the non-AZM treatment arms and the AZM treatment arms (HCQ vs HCQ + AZM, CQ vs CQ + AZM). If either of the AZM arms have significantly faster recovery rates (two-sided *P*-value < 0.05) compared to the corresponding non-AZM arm group, then we will conclude that the AZM group is superior to the non-AZM group. We will also compare recovery rates between the HCQ vs CQ, and HCQ + AZM vs CQ + AZM treatment arms to assess whether HCQ and CQ treatments differentially influence recovery. We will then proceed to a multivariable Cox-regression model that adjusts symptom severity at baseline (sum of presence of fever ≥ 38.0°C and total number of 'Quite a Bit' or 'Very much' responses to the Flu-PRO questionnaire [maximum total = 20]). The final model will retain the treatment groups and those covariates that are significant ($P < 0.05$) in the multivariable model and we will report the model-adjusted hazard ratios and 95% CI for the treatment groups as well as the covariates. Participants that are lost to follow-up or die will be censored at the date of last contact or date of death, respectively, and patients that do not recover during the study period will be censored at 42 days. The primary analysis will be intention-to-treat and a per-protocol analysis will also be performed as a secondary analysis of the primary endpoint. Statistical analyses will be conducted using PROC LIFETEST and PROC PHREG as appropriate (SAS Institute Inc, Cary, NC, USA).

Sensitivity analyses
In our primary analysis death is treated as a competing risk for recovery (i.e., someone that dies will never recover) by censoring subjects at their date of death. Although we anticipate low mortality rates (e.g., 1.4%[2]) censoring patients due to death has the potential to bias recovery rate estimates[8]. In order to assess this assumption we will explore the use of a more complex approach that explicitly models the subdistribution of recovery rate[9]. If results from this analysis are substantially different form the primary analysis, then we will use model fit statistics and Monte Carlo simulation to determine whether the more complex model better describes the data generating process.

Secondary and exploratory endpoint analyses
Secondary endpoints include time to resolution of fever, time to improvement of symptoms, duration of hospitalization, proportion requiring supplementary oxygen above baseline usage, duration of requirement for supplementary oxygen, proportion requiring ICU admission, proportion requiring mechanical ventilation, ventilator free days, all-cause mortality. Exploratory COVID-19 specific mortality, number of patients with diagnosed cardiomyopathy, number of patients with new onset arrhythmia. All secondary and exploratory endpoints will be analyzed by survival analysis (KM and/or Cox), t-tests, chi-square tests, or Fisher's exact tests as appropriate.

Sample size and power analysis

A recent COVID-19 study in the Jiangsu province of China[10] indicated that fever in about half of COVID-19 infected people lasted less than 7 days. Because this research area is rapidly developing and there is limited knowledge about this disease, we use the simple median estimate of 7-days as a starting point for our recovery outcome. In addition to attenuation of fever, our primary outcome adds criteria that fever must be attenuated for at least 48 hours and there needs to be symptom improvement. Given our criteria we assume 11 days as a rough estimate for median recovery time in the non-AZM treatment groups to compare the AZM treatment groups against in our study. This study is powered to test the hypothesis that HCQ + AZM improves recovery rate compared to HCQ alone, and CQ + AZM improves recovery rate compared to CQ alone. Assuming a minimum improvement of 4 days in the median recovery time (median recovery time of 7-days) and a censoring rate due to death or loss to follow-up of 0.04 per day, we plan on enrolling a total of 500 participants (125 per arm) which would give us just over 80 percent power ($1 - \beta = 82\%$, $\alpha = 0.05$) to test our primary hypotheses.

SOURCE DOCUMENTATION

All source documents will be kept in an appropriately secure office in a locked high density filing system in a locked office.

QUALITY CONTROL

All eligibilities and ICFs and a portion of the study case report forms (no less than 10%) will be audited by the study quality management personnel.

ETHICS AND PROTECTION OF HUMAN SUBJECTS

Once the study is approved by the Washington University Human Research Protection Office (HRPO), recruiting and screening will begin for the study. All subjects or their legally authorized representative (LAR) are required to read English, understand and sign the informed consent document.

DATA HANDLING AND RECORD KEEPING

Data will be generated and managed according to Washington University School of Medicine and HIPAA policy.

Data will be entered onto a central Redcap database housed at Washington University. All research records will be maintained under the two-lock rule with access limited to authorized study personnel or regulatory authorities. The study records will be maintained up to seven years after the completion of the study.

PUBLICATION POLICY

The PI and co-investigators will participate in manuscript preparation and submission to journals.

LITERATURE REFERENCE

1. Onder G, Rezza G, Brusaferro S. Case-Fatality Rate and Characteristics of Patients Dying in Relation to COVID-19 in Italy. *JAMA.* 2020.

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4. Liu J, Cao R, Xu M, et al. Hydroxychloroquine, a less toxic derivative of chloroquine, is effective in inhibiting SARS-CoV-2 infection in vitro. *Cell Discov.* 2020;6:16.
5. 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;14(1):72-73.
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8. Pfirrmann M, Hochhaus A, Lauseker M, Saussele S, Hehlmann R, Hasford J. Recommendations to meet statistical challenges arising from endpoints beyond overall survival in clinical trials on chronic myeloid leukemia. *Leukemia.* 2011;25(9):1433-1438.
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10. Xu T, Chen C, Zhu Z, et al. Clinical features and dynamics of viral load in imported and non-imported patients with COVID-19. *Int J Infect Dis.* 2020.

Appendix 1 Flu-PRO questionnaire (Will be modified to read COVID-19 where Flu/influenza referenced)

Participant ID:_____Participant Initials:_____Date:____/_____/_____

~~Influenza~~ Symptoms Questionnaire

People experience ~~the Flu~~ COVID-19 in different ways. We would like to know about the symptoms you have been experiencing during the last 24 hours. For each symptom, please mark one box under the response that best matches your experience. Mark the "Not at all" box, if you did not have that symptom in the past 24 hours.

What time is it? _____ AMP / PM (please circle)

Please rate the extent which you had each symptom during the past 24 hours.

Symptom	Not at all	A little bit	Somewhat	Quite a bit	Very much
Runny of dripping nose	☐	☐	☐	☐	☐
Congested or stuffy nose	☐	☐	☐	☐	☐
Scratchy or itchy throat	☐	☐	☐	☐	☐
Difficulty swallowing	☐	☐	☐	☐	☐
Teary or watery eyes	☐	☐	☐	☐	☐
Sore or painful eyes	☐	☐	☐	☐	☐
Eyes sensitive to light	☐	☐	☐	☐	☐
Trouble breathing	☐	☐	☐	☐	☐
Chest congestion	☐	☐	☐	☐	☐
Chest tightness	☐	☐	☐	☐	☐
Dry or hacking cough	☐	☐	☐	☐	☐
Wet or loose cough	☐	☐	☐	☐	☐
Headache	☐	☐	☐	☐	☐
Head congestion	☐	☐	☐	☐	☐
Sinus pressure	☐	☐	☐	☐	☐
Felt dizzy	☐	☐	☐	☐	☐
Felt lightheaded	☐	☐	☐	☐	☐

Appendix 2 Contraindicated* with study drugs (predominantly chloroquine)

Type	Medication
Analgesic	methadone
Antiarrhythmic	amiodarone, digoxin, disopyramide, dofetilide (*Tikosyn*), dronedarone (*Multaq*), flecainide, ibutilide (*Corvert*), procainamide, quiNIDine, sotalol
Antibiotic	bedaquiline, clarithromycin, delamanid, rifampin
Anticonvulsant	carbamazepine, phenytoin
Antidepressant	bupropion, fluoxetine, paroxetine
Antifungal	itraconazole, ketoconazole, posaconazole, voriconazole
Antihormone therapy	abiraterone, apalutamide, enzalutamide,
Antimalarial	artemether/lumefantrine, dapsone, quiNINE, mefloquine
Antineoplastic	ceritinib (*Zykadia*), dabrafenib (*Tafinlar*), idelalisib, ivosidenib (*Tibsovo*), lenvatinib (*Lenvima*), mitotane, vandetanib (*Caprelsa*)
Antipsychotic	chlorproMAZINE, ziprasidone
Antiviral	cobicistat, darunavir, lopinavir, nelfinavir, ritonavir, saquinavir, tipranavir
Immunosuppressant	CycloSPORINE
Misc. injection/intravenous	agalsidase α or β, conivaptan

*Includes interactions severe enough to require constant monitoring during co-administration (Category D)