Intensive Care Unit, Pharmacy Department and Infectious Diseases Department,

Royal Brisbane & Women's Hospital

and

University of Queensland Centre for Clinical Research, The University of Queensland

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and

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Use of therapeutic drug monitoring (TDM) to optimise oral/enteral hydroxychloroquine dosing in critically ill patients with COVID-19

PROTOCOL VERSION: 1.0

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Contents

1. List of Abbreviations

ADR	Adverse drug reaction
AE	Adverse event
ALT	Alanine aminotransferase
APACHE	Acute Physiology & Chronic Health Evaluation
BMI	Body mass index
CI	Chief Investigator
CK	Creatinine kinase
Co-I	Co-investigator
CRP	C-reactive protein
CRRT	Continuous renal replacement therapy
DOB	Date of Birth

EDC	Electronic data capture
EDTA	Ethylenediaminetetraacetic acid (an anticoagulant)
EUC	Electrolytes, urea & creatinine
CRF	Case report forms
FBC	Full blood count
GCP	Good clinical practice
GFR	glomerular filtration rate
GP	General practitioner
HCQ	Hydroxychloroquine
HRN	Hospital record number
HREC	Human research ethics committee
ICH	International Conference on Harmonisation
ICU	Intensive care unit
ID	identification
ID physician	Infectious disease physician
IV	Intravenous
IVI	Intravenous infusion
LFT	Liver function test
LiHep	Lithium heparin
MIC	Minimum inhibitory concentration
NHMRC	National Health and Medical Research Council
NOK	Next of kin
PBPK	Physiology based pharmacokinetics
PK	Pharmacokinetic
PI	Principal Investigator (a clinician responsible for one site)
QALY	Quality-adjusted life years
QID	4 times per day
RC	Research co-ordinator
RCT	Randomised control trial
SAE	Serious adverse events
SOP	Standard Operating Procedure
TDS	3 times per day
TDM	Therapeutic drug monitoring
TGA	Therapeutic Goods Administration
UQCCR	University of Queensland Centre for Clinical Research

2. General Information

2.1. Title

Use of therapeutic drug monitoring (TDM) to optimise oral/enteral hydroxychloroquine dosing in critically ill patients with COVID-19

2.2. Data Management Centre & Responsible Institution

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2.3. Investigators

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3. Background Information

3.1. Rationale

As of March 27, 2020, the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has resulted in nearly 481,400 confirmed infections and almost 22,000 deaths globally. SARS-CoV-2 is a betacoronavirus belonging to a unique clade of the sarbecovirus subgenus of the Orthocoronavirdinae subfamily first identified in January 2020[1]. Initial reports suggest that infection is associated with severe disease requiring intensive care admission in ~5% of proven infection[2]. Risk factors for requiring intensive care support include age and comorbid conditions, commonly cardiac disease and diabetes[3]. Median duration between onset of symptoms and ICU admission is typically 9-10 days with acute respiratory distress syndrome (ARDS) being the main reason for needing intensive care support[4]. ICU admission for COVID-19 (disease caused by SARS-CoV-2 infection) was associated with a 61.5% mortality in one study[5]. Currently, there are no proven effective therapeutic options to reduce the mortality for SARS-CoV-2 infection. A small randomised controlled trial by Cao *et al* evaluating the use of lopinavir-ritonavir in patients hospitalised with COVID-19 revealed no demonstrable clinical benefit with its use but was underpowered to detect a clinically significant difference[6].

Many agents have shown promise in inhibiting coronavirus replication *in vitro*[7]. Previous studies undertaken for SARS-CoV and MERS-CoV have provided an opportunity to accelerate the identification of efficacious therapies. Remdesivir (GS-5734), a nucleotide analogue prodrug (currently in clinical trials for Ebola virus infection) and an old HIV-1 protease combination lopinavir/ritonavir (LPV/r) have shown favourable results against coronaviruses in vitro and in vivo. Unfortunately remdesivir is not widely available and concerns over drug toxicity, drug interactions and efficacy with use of LPV/r have been raised[6]. Chloroquine has also shown great promise in treating coronaviruses in preclinical studies[8]. Although not licensed for clinical use in Australia, chloroquine is a widely used anti-malarial drug with immunomodulatory effects and has been shown to inhibit growth of SARS-CoV-2 *in vitro*. In order for many viruses to replicate within cells they require endosomal/lysosomal acidification and acidic pH dependent endosomal proteases to cleave the viral glycoprotein segments. Without these processes functioning, viral replication is halted. Chloroquine, and its structural analogue hydroxychloroquine, is reported to rapidly cross cellular membranes and inhibit viral infection by increasing endosome/lysosome pH[9]. At a cellular level in tissues, it is able to become highly concentrated in acidic organelles leading to dysfunction of several enzymes required for proteolytic processing and post-translational modification of viral proteins. In addition, this may inhibit synthesis of many cytokines, chemokines or mediators which contribute to severity of viral illness. Hydroxychloroquine was found to have more potent activity against SARS-CoV-2 than chloroquine in one study[8]. In addition, a non-randomised controlled trial using hydroxychloroquine plus azithromycin as treatment of SARS-CoV-2 infection in 20 patients reported significant reduction in viral loads and improvement in patient symptoms (Gautret et al 2020 IJAA in press), although this small study is not without limitations. At least 10 clinical trials are currently underway or planned to assess chloroquine or hydroxychloroquine efficacy[10].

There are a number of trials currently recruiting or planned assessing different treatment strategies for SARS-CoV-2 infection. In Australia, the ASCOT trial plans to recruit more than 2200 non-ICU

patients into one of four treatment arms (1) standard of care, (2) LPV/r, (3) hydroxychloroquine, or (4) LPV/r + hydroxychloroquine. Currently there is no nationwide strategy for how ICU patients with severe SARS-CoV-2 infection will be managed.

Along with pre-clinical data from animal models, there are emerging data from *in vitro* cell culture models which provide some data of the anti-viral effects of tested compounds. Application of sophisticated *in silico* modelling and simulation techniques is then able to advance infection model-defined exposure targets into humanised doses. Whilst this process provides valuable guidance for anti-viral choice, these methods are analogous to those applied in the drug development process and only propose doses that then require human/clinical validation. The weaknesses of these pre-clinically validated dosing regimens, in part, relate to the relevance of the available pharmacokinetic data for the drug which is applied in the simulation process. That is, pharmacokinetic data that is not from the population of interest, for example data from healthy volunteers as opposed to severely ill patients with active infection and respiratory dysfunction, may have different bioavailability (for orally or subcutaneously administered drugs), volume of distribution and drug clearance which means that such doses may be sub-optimal.

Drugs for SARS-COV-19 may also introduce other treatment challenges, particularly in terms of drug interactions. Various compounds that have been proposed for treatment of SARS-COV-19 are affected by the CYP450 metabolising system as either substrates, enzyme inhibitors or enzyme inducers and consideration of these effects on dosing requirements of concomitant SARS-COV-19 or other supportive drug therapies.

The Royal Brisbane and Women's Hospital Intensive Care Unit clinical Consultant Medical Staff, in agreement with the Infectious Diseases Department and the Pharmacy Department, have decided that all patients admitted to the ICU should receive hydroxychloroquine as standard of care. Therefore, hydroxychloroquine is NOT considered a trial intervention for the purpose of this study. Hydroxychloroquine has been chosen in preference to chloroquine due to drug availability in Australia.

To our knowledge, every other hospital using hydroxychloroquine for Covid-19 infection as either standard care or as part of a clinical trial is using a single dose for all patients, extrapolated from use as an antimalarial in patients who are not critically ill. Given the considerable uncertainty about the effect of pharmacokinetic variability on dosing requirements in individual Covid-19 patients who are critically unwell, at the Royal Brisbane and Women's Hospital we will instead support the proposed dosing with therapeutic drug monitoring (TDM) to ensure we can adapt dosing to concentrations that are likely to effective. The TDM assays will be performed by Department of Chemical Pathology, Pathology Queensland.

The research question posed by this study is, therefore, whether the application of TDM to hydroxychloroquine dosing in Covid-19 patients results in clinically significant alterations in the drug dosing regimen. We hypothesise this will be the case. If so, the findings will have profound implications for the conduct and interpretation of several very large clinical trials currently in their early stages worldwide.

3.2. Quality Improvement Activity

RBWH has a 27-bed medical and surgical ICU that is expected to at least double in capacity with the expected imminent need. It is expected that this surge in severe cases may occur as early as the first

week of April 2020. Although studies are underway, there is no current proven therapy to reduce mortality associated with COVID-19. However, given the severity of disease and risk for death, based on an informal survey of intensivists it is expected that hydroxychloroquine therapy will be offered as a standard to severe cases of COVID-19 infection admitted to ICU at RBWH. While this therapy holds promise, its safety and efficacy have not been clearly established for this purpose. Therefore, this protocol will monitor the safety of use of hydroxychloroquine for management of severe COVID-19 infection. As data becomes available on clinical outcomes with other agents currently being assessed in other trials, this standard of care may change.

4. Aim, hypothesis and enabling objectives

4.1 Aim

The aim of the study is to define the dosing of hydroxychloroquine required to achieve a therapeutic drug level in patients with COVID-19 and critical illness.

4.2 Hypothesis

Patients with COVID-19 and critical illness will require adjustment of standard hydroxychloroquine doses in order to achieve safe therapeutic levels over a treatment course of up to 10 days while receiving treatment in an ICU.

4.3 Enabling objectives

- To monitor the safety and describe the dosing and characteristics of TDM performed for hydroxychloroquine (HCQ) in patients admitted to the RBWH ICU with severe COVID-19.
- To monitor clinical outcomes of intensive care patients with confirmed SARS-CoV-2 infection treated with HCQ

5. Design

This is an investigator-initiated single-arm, non-randomised open label quality improvement study designed to ensure the safe and optimal dosing of HCQ in critically ill COVID-19 patients using TDM in the first 10 days after commencement of HCQ in an ICU. The clinical outcomes and complications of ICU patients treated with HCQ for severe COVID-19 will be monitored.

6. Endpoints

6.1. Primary endpoints

Dose adjustment required to attain a steady-state target trough hydroxychloroquine concentration of 0.24mg/L using TDM (proportion of patients / magnitude of adjustments)
Timepoint: Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose

6.2. Secondary endpoints

1. Achievement of free HCQ concentrations >0.24 mg/L after dose adjustment using TDM. (proportion of patients / magnitude of excess) / Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose.
2. 30-day case-fatality following ICU admission / 90 days after ICU admission
3. ICU case fatality / 90 days after ICU admission
4. Hospital case fatality / 90 days after ICU admission
5. Change of worst daily SOFA score / Daily up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose
6. Time to negative SARS-CoV-2 PCR results / Up to 90 days after ICU admission
7. Change in routine blood test results (as available) / Daily up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose
8. Routine chest imaging results (as available) / Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose
9. Hydroxychloroquine pharmacological data including drug dose and dosing history, formulation used (tablet, capsule, liquid), administration route (swallowed whole, liquid, crushed), concomitant medication / Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose
10. Worst daily PaO2 / FiO2 / Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose
11. All antibiotics used each day / Up to the point of ICU discharge, in the first 10 days after commencement of initial HCQ dose

7. Selection of Participants

7.1. Inclusion criteria

1. Suspected or proven severe COVID-19 infection admitted to the ICU.
2. Prescribed hydroxychloroquine by the treating clinician as part of routine management for COVID-19 infection

7.2. Exclusion criteria

1. Age <18 years old

8. Assessment of Participants

8.1. Procedures

8.1.1.Current practice for dosing HCQ

Hydroxychloroquine is an old antimalarial drug which has different dosing regimens for the various malaria and auto-inflammatory indications.

It will be the RBWH ICU policy that standard dosing for HCQ for severe COVID-19 will be 400mg twice daily for days 1-2 followed by 200mg twice a day for up to 8 days (maximum 10 days total)

An ECG will be done at baseline, day 2 and day 7 of HCQ therapy.

8.1.2.Current practice for HCQ dose optimisation for severe COVID-19

As per standard and current routine clinical practice at the RBWH ICU, TDM is led by the Pharmacist Consultant/senior ICU pharmacist after initiation of therapy. As per this practice, trough venous blood samples (3 ml) will be taken via an indwelling arterial cannula. The first TDM should ideally occur before the administration of the FIRST MAINTENANCE DOSE, provided the appropriate loading dose schedule has been completed. Sampling thereafter can occur every 24-48 hours to confirm the appropriateness of ongoing dosing as guided by the Pharmacy and Medical Team. Blood samples will be transported to Queensland Pathology on the Herston Campus for drug analysis. Doctors and pharmacists will be able to fill out the pathology request form for HCQ concentrations.

The Metavision clinical information system for the RBWH ICU will have patient record entries for request for TDM and interpretation of TDM results inputted by clinical pharmacists. Prescribing of initial and ongoing/revised dosing will remain the responsibility of medical officers.

Samples for assay will be spiked with stable isotope labelled internal standards and drugs extracted by liquid-liquid extraction with tertbutyl methyl ether. Extracts will be analysed by a validated UHPLC-MS/MS chromatographic method to measure total drug concentrations. Test samples will be assayed alongside calibrators and quality controls and be subjected to industry standard batch acceptance criteria.

A pharmacogenetic evaluation of CYP3A4 and CYP2D6 expression will be performed on existing blood samples to ascertain information regarding HCQ metabolism and drug exposure.

In order to further interrogate the collected data and scientifically evaluate the appropriateness of hydroxychloroquine, as a secondary analysis, we will analyse the dosing and concentration data of 100 patients and subject this to population pharmacokinetic modelling using non-parametric pharmacokinetic modelling approach in Pmetrics®. The final pharmacokinetic population parameters estimated from all possible covariates will be chosen based on the value of mean weighted error (describing the precision), mean weighted squared error (describing the bias), and coefficient of determination (r^2). Monte Carlo simulations will be performed to identify the most optimal dosing regimens that will target drug concentrations.

8.1.3.Target exposure

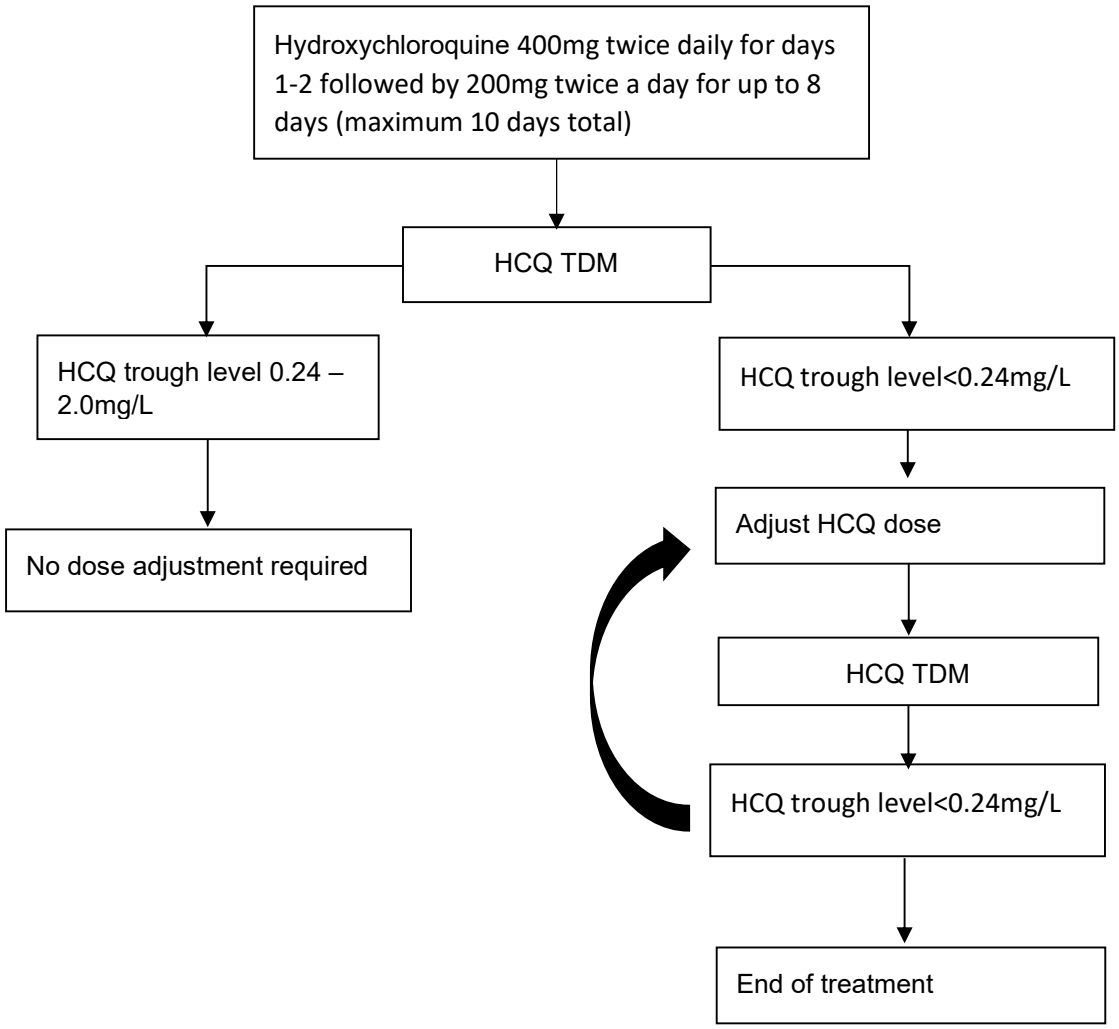
Primary target endpoint exposure will be defined as steady-state hydroxychloroquine trough concentrations of 0.24mg/L.

8.1.4.Other data collection

The following parameters that are routinely collected as part of standard practice in the ICU will also be reported on in the quality improvement activity

1. Demographic data (age, gender, height, weight, BMI)
2. ICU admission APACHE II, APACHE III
3. Evidence of specific end-organ dysfunction
4. Fluid balance
5. Biochemistry (albumin, liver function, serum creatinine)

Figure 1. Current practice of HCW dose optimisation at RBWH ICU



9. Criteria for discontinuing individual patient study participation

Patients will remain part of the TDM study until the soonest of:

- the time they are discharged from the ICU (either alive or dead);

- day 10 after the commencement of the first HCQ dose; or
- the time at which their treating clinician permanently discontinues the administration of HCQ for any reason.

After discontinuation of study participation, patient outcomes will be ascertained up to 90 days following the first HCQ dose.

10. Duration of Data Collection

Pharmacokinetic, physiologic and other detailed patient data will be recorded from time of ICU admission until ICU discharge or until day 10 after ICU admission, whichever is the sooner. Following ICU discharge, patient outcomes will be censored at hospital discharge, 30 days or 90 days, as listed in section 5.2.

11. Termination of Activity

The quality improvement activity may be terminated at any time at the request the Chief Investigator, or a regulatory authority, with proper and timely notification of all parties concerned.

12. Procedures

12.1 Case report forms

The investigators will document HCQ TDM values and safety data. All other clinical and physiological data variables will be accessed from the Meta-vision Clinical Information System either contemporaneously or retrospectively. Required data described in the protocol will be available in the patient medical record.

12.2 Data collection

Data collection will be by paper files and the Meta-vision Clinical Information System with the quality improvement activity site taking responsibility for patient identification as well as patient sampling and data collection.

13 Statistics

13.1 Sample size

The aim of the study is to define the dosing of hydroxychloroquine required to achieve a therapeutic drug level in patients with COVID-19 and critical illness. The number of patients required to achieve this will be determined by the variability of the enrolled population's drug levels. As this is not possible to know in advance, sample size is not possible to define.

13.2 Justification

Based on previous physiologically based pharmacokinetic studies, using a target HCQ trough concentration of 0.24mg/L will maximise lung exposure to drug. We will do this using a non-parametric pharmacokinetic modelling approach in Pmetrics®. Experience of this approach in this population with other drugs suggests a sample size of 100 will be sufficient to account for likely variability, but the study will continue until a robust population pharmacokinetic model taking into account organ dysfunction and other patient characteristics has been defined.

13.3 Statistical methods

Analysis will be primarily descriptive. The cumulative incidence of primary and secondary endpoints will be reported.

14 Quality Control and Quality Assurance Monitoring

14.1 Responsibilities of the Chief Investigator

The Chief Investigator agrees to perform the quality improvement activity in accordance with this protocol, the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and the applicable regulatory requirements. The Chief Investigator is required to ensure compliance with all procedures required by the protocol. This protocol will be approved by the Department of Intensive Care Services Quality improvement program, RBWH prior to commencement.

15 Ethical Considerations

15.1 Ethical Principles

This quality improvement activity will be conducted in accordance with the principles laid down by the ICH guidelines for Good Clinical Practice, the Australian National Health and Medical Research Council National Statement on Ethical Conduct in Human Research (2007, updated 2018) and all applicable local regulatory requirements.

15.2 Waiver of full ethical review

Based on the National Statement on Ethical Conduct in Human Research, the Chief Investigator will seek a waiver of full ethical review for this quality improvement activity.

Specifically, the Chief Investigator considers that:

1. The quality improvement activity involves no more than minimal risk to the participants as all ICU patients admitted with severe SARS-CoV-2 infection will be treated with HCQ as part of what has been agreed to be best-practice treatment in the Royal Brisbane and Women's Hospital. TDM of other drugs is routinely employed in this hospital. All patients involved in this activity will receive the same dose adjustment strategy, the primary objective of which is to ensure the safety and effectiveness of what has been agreed a standard care. The only intervention proposed is data collection to quantify the effectiveness of this approach.

2. A waiver of informed consent will not adversely affect the rights and welfare of these patients.
3. There is sufficient protection of patient privacy and an adequate plan to protect the confidentiality of collected data at all times.

16 Safety

HCQ is not considered an investigational drug for the purpose of this study, and so Adverse Events, Serious Adverse Events and Suspected Unexpected Serious Adverse Drug Reactions will not be reported in the manner that is usually the case for a clinical trial of a pharmaceutical agent.

17 Data Handling and Record Keeping

A guide to the data collection with definitions and rationale will be provided together with a paper version of the data collection forms. Paper documents will be stored in secure locked cabinets with access limited to authorised persons. The principle means of data collection and data processing will be via original paper forms and the data in the Meta-vision Clinical Information System**.** All forms will be signed and dated by the authorised research staff and all changes made following data submission will be recorded. When archiving or processing data pertaining to the investigator and/or to the patients, the coordinating centre shall take all appropriate measures to safeguard and prevent access to this data by any unauthorised third party. The Chief Investigator is responsible for maintaining all documentation of quality improvement activity confidential, and take measures to prevent accidental or premature destruction of these documents. The Chief Investigator is responsible for retaining the documents for at least fifteen years after the completion or discontinuation of the quality improvement activity.

18 Financing and Insurance

All costs related to ethics preparation, patient enrolment, data collection and sample collection assaying will be incurred as part of standard clinical care (no additional costs) with costs related to data analysis incurred by The University of Queensland. As a Quality Improvement Project of the Royal Brisbane and Women's Hospital, indemnity will be provided by MetroNorth Hospital and Health Service of Queensland Health.

19 Registration

As a clinical study investigating the hypothesis that TDM will influence dosing of HCQ, this activity will be prospectively registered on the Australian and New Zealand Clinical Trials Registry.

20 Project Timeline

Date	**Project Milestone**

March 2020	Quality Improvement Activity Protocol and forms finalised
March to December 2020	Data Collection
January to March 2020	Database lock, data analysis and initial results

21 References

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