Original Manuscript

Title: Repositioning chloroquine as an ideal antiviral prophylaxis against COVID-19 – Time is now

Raymond Chang MD[1], Wei-Zen Sun MD[1,2]

[1]Institute of East-West Medicine, New York, USA; chang@meridianmedical.org

[2]National Taiwan University Hospital, Taipei, Taiwan; wzsun@ntu.edu.tw

Corresponding author: Raymond Chang : chang@meridianmedical.org

Abstract

The novel coronavirus 2019 (COVID-19) pandemic is rapidly advancing despite public health measures. Pharmaceutical prophylaxis is an established approach to potentially control infectious diseases and is one solution to the urgent public health challenge posed by COVID-19. Screening and development of new vaccines and antivirals is expensive and time consuming while the repositioning of available drugs should receive priority attention as well as international government and agency support. Here we propose an old drug chloroquine (CQ) to be urgently repositioned as an ideal antiviral prophylactic against COVID-19. CQ has ability to block viral attachment and entry to host cells. Its proven clinical efficacy against a variety of viruses including COVID-19 and its current deployment in COVID-19 therapeutic trials strengthens its potential candidacy as a prophylactic. Furthermore, CQ has a long safety record, is inexpensive and widely vailable. Here we reviewed CQ's antiviral mechanisms, its laboratory efficacy activity against COVID-19, as well as CQ's pharmacokinetics in its established use against malaria and autoimmune diseases to recommend safe and potentially efficacious dose regimens for protection against COVID-19: a pre-exposure prophylaxis of 250-500mg daily and post-exposure prophylaxis at 8mg/kg/day for 3 days. We recommend further urgent research on CQ for COVID-19 prevention and urge that the above regimens be investigated in parallel with mass deployment by relevant agencies in attempts to contain the pandemic without unnecessary regulatory delays as benefits far outweigh risks or costs.

1. Introduction

Since its reported outbreak in late 2019 (Zhu et al., 2020), the corona virus 2019
(COVID-19) has exploded from a few people suffering a respiratory disease in the
Chinese city of Wuhan to a pandemic of over 100,000 cases with thousands of deaths.
Current methods of pandemic control is confined only to public (travel restrictions,
quarantines, avoidance of gatherings, school closures) and personal (face mask use,
hand hygiene) health measures, while vaccine development will cost billions of
dollars and maybe as much as 18 months away from deployment (Kuchler et al.,
2020).

Pharmaceutical antivirals are not only potentially therapeutic but have been
successfully applied pre- and post-exposure as prophylaxis against viral infections
such as influenza (Oxford, 2007), human immunodeficiency virus (HIV) (Desai et al.,
2017), cytomegalovirus (Hussein et al., 2020), and respiratory syncytial virus (Rezaee
et al., 2017). Using influenza as a model for preventive management of respiratory
viral pandemics, the key concerns are surges in community attack rates and
healthcare system demand (Nap et al., 2007) , which in turn lead to disruptions in
healthcare with potentially disastrous social and economic ramifications. In their
systemic review and meta-analysis of effective interventions to contain an influenza
pandemic, Saunders-Hastings et al. identified vaccination and antiviral prophylaxis as
two major pharmaceutical interventions that can be effective (Saunders-Hastings et
al., 2016). However, to date we have neither developed a vaccine nor is there any
approved or established antiviral prophylaxis in deployment against COVID-19.

In the case of COVID-19, hiding in plain view is a plausible and potential
prophylaxis option that can be relatively easily achievable by repositioning the old
drug chloroquine (CQ), one of the most prescribed drugs in the world today (White et

al., 2014). CQ has been long used as chemoprophylaxis against malaria and has known antiviral properties. Although largely taken over by newer and more effective agents, CQ is a drug that has been in use for over half a century as a chemoprophylactic agent against malaria (Peters, 1971) and still in use today.

This article sets out to review the relevant experimental results of CQ as an antiviral as well as its pharmacokinetic (PK) data and its toxicities to suggest that CQ is an ideal candidate that should be urgently repositioned as an antiviral prophylactic against COVID-19.

2. Background on CQ

CQ is a 4-aminoquinoline that is most well-known as an anti-malarial. It was originally discovered in 1934 and its full clinical development involved investigators from six countries on five continents over a decade before clinical trials confirmed its therapeutic value as an anti-malarial drug (Coatney, 1963). It was clinically introduced as a prophylactic treatment of malaria in 1947 and subsequently included by the World Health Organization in its model list of essential medicines which includes drugs deemed essential in addressing the most important public health needs globally. In the United States, chloroquine is FDA-approved for the treatment and prophylaxis of uncomplicated malaria in countries where chloroquine-sensitive malaria is present and the treatment of extra-intestinal amebiasis. Besides its anti-malarial properties, CQ also has established immunomodulatory and anti-inflammatory effects (Al-Bari, 2015) and current non-FDA approved or repositioned use of CQ include the potential treatment of a wide spectrum of diseases, both non-infectious and infectious such as a range of cancers (Manic et al., 2014), rheumatoid arthritis, systemic lupus erythematosus, systemic sclerosis, primary progressive

multiple sclerosis, Q fever, Whipple's disease, and a variety of fungal and viral infections, (Plantone & Koudriavtseva, 2018).

2.1 CQ as antiviral – *in vitro* and *in vivo* studies

CQ's bioactivity against viruses have been reported a half-century ago (Shimizu et al., 1972) and it's potential to be repositioned as a broad-spectrum anti-microbial against bacteria, fungal and viral infections was proposed over a decade ago (Rolain et al., 2007).

CQ has direct and indirect anti-viral effects. Direct antiviral activity of CQ has been identified against a range of no less than thirty viruses mostly by *in vitro* studies (Rolain et al., 2007). The mechanisms of direct inhibition by impeding viral entry as well as disrupting post-entry viral envelope maturation by CQ has been reviewed (Savarino et al., 2003). It has been subsequently demonstrated that CQ targeting of endosomal acidification and resultant alkalinization of cellular organelles and inactivation of pH-dependent enzymatic processes impedes viral entry as well as replication and is the basis of its potential as an antiviral (Al-Bari, 2017).

Upon attachment to cells, a virus needs to fuse to the host o cell to deliver the viral genome. Preventing viral entry by inhibiting attachment and fusion are ideal for prophylaxis against infection. This approach has been successful with HIV and has been demonstrated to be viable *in vitro* with CQ against the Ebola (EBOV), influenza and Marburg viruses (Long et al., 2015).

Another direct antiviral mechanism of CQ involves impairment of pH-dependent protease and glycosyltransferase enzymes in the endoplasmic network needed for post-entry viral envelope maturation, which has been demonstrated in

experiments with Flaviviruses (Randolph et al., 1990), Dengue (DENV) and Chikungunya (CHIKV) viruses.

Besides acting directly on the virus, there are possible indirect antiviral effects that impede viral cellular entry and infection. For example, CQ has been demonstrated to interfere with terminal glycosylation of the cellular receptor angiotensin-converting enzyme 2 (ACE2) which facilitates entry of severe acute respiratory syndrome corona virus (SARS-CoV) thus potentially reducing virus-receptor binding and abrogating infections (Vincent et al., 2005). Significantly, SARS-CoV is also an animal derived human corona virus (HCoV) in the same *sabrecovirus* subgenus of the c*oronaviridiae* virus family as COVID-19 which shares the ACE2 pathway to initiate an infection (Hoffmann et al., 2020). Separate CQ studies with SARS-CoV showed significant prophylactic (Vincent et al., 2005) and post-infection (Keyaerts et al., 2004) activity, with cell culture studies demonstrating CQ's effectiveness in preventing infection if the drug is added 24 hours prior to infection and even if added 5 hours post infection (Vincent et al., 2005). Besides SARS-CoV, CQ also demonstrated antiviral activity against five out of seven known human corona viruses including COVID-19 (Wang et al., 2020), MERS-CoV (De Wilde et al., 2014), HCoV-229E (Kono et al., 2008) , and HCoV-OC43 (Keyaerts et al., 2009). In the case of COVID-19 as in SARS-CoV, time-of-addition assay demonstrated that CQ functioned at both entry and at post-entry stages of infection in the VERO E6 cells assay used (Wang et al., 2020).

Specifically, Wang reported the 50% effective concentration (EC_{50}) of CQ against COVID-19 using infected VERO E6 cells as determined by CCK8 assay to be 1.13 µM and the EC_{90} was 6.90 µM, indicating potent viral inhibition at micromolar concentrations (Wang et al., 2020). For comparison with activity against other

HCoVs, the EC_{50} was 3.6 μM for MERS-CoV (De Wilde et al., 2014), between 2.3

μM (De Wilde et al., 2014) to 4.4 μM (Vincent et al., 2005) for SARS-CoV and 0.3

μM for HCoV-OC43 replication in HRT-18 cells (Keyaerts et al., 2009).

In animal studies, CQ can prevent DENV infection in Aotus monkeys (Farias

et al., 2015), reduce zika virus induced mortality when administrated soon after

infection (C. Li et al., 2017), protect mice against a deadly challenge dose of EBOV

(Madrid et al., 2013), and reduce mortality of lethal human coronavirus HCoV-OC43

infection in newborn C57BL/6 mice when CQ was acquired through the placenta or

via maternal milk (Keyaerts et al., 2009).

2.2 CQ as antiviral - clinical studies

There has only been a few small clinical studies to date using CQ clinically against

viral infections. In HIV, the CQ derivative hydroxychloroquine (HCQ) at 800mg

daily for eight weeks was found to have a 0.6 $\log_{10}$ reduction of HIV-1 load (P =

0.022) when compared to untreated controls (Sperber et al., 1995). In a study on

biopsy proven chronic active hepatitis B who received 50–450 mg of CQ for a median

of 12 months normalized their alanine aminotransferase (ALT) (Kouroumalis &

Koskinas, 1986). Other trials investigated the antiviral effects of CQ for 3 days

beginning 72 hours after infection by DENV and demonstrated CQ reduction of

occurrence of dengue hemorrhagic fever as well as decrease patients' perceived

intensity of pain and improve their daily activity performance (Tricou et al., 2010).

Perhaps most significantly, Chinese researchers just published a breakthrough

interim report on an ongoing multicenter controlled trial involving more than ten

hospitals using CQ as treatment for COVID-19 and results are encouraging. Results on

over a hundred patients so far have demonstrated that CQ phosphate at 500mg twice

daily for 10 days is superior to the control, without serious adverse reaction and has prevented the exacerbation of pneumonia, improved lung imaging findings, promoted viral seroconversion, and reduced clinical duration of disease meaningfully (Gao et al., 2020).

2.3 CQ – pharmacokinetics & pharmacology considerations

Pharmacokinetically, CQ is rapidly and well absorbed orally with good bioavailability (>75%) and peak serum levels is achieved within 2-3 hours. Approximately 55% of the drug in the plasma is bound to non diffusible plasma constituents. It undergoes primarily hepatic metabolism by cytochrome P450 enzymes and has a very long plasma terminal elimination half-life of 1-2 months and after a single dose the drug can be found in the liver and urine for up to five years. The long half-life reflects its high volume of distribution of greater than 100L/kg which extends into aqueous compartments and with about half the metabolites undergoing renal clearance (Krishna & White, 1996). Significantly for potential use against a respiratory virus, peak tissue/plasma concentration ratio greater than 300 is obtained in many tissues including lungs, and the concentration increased with chronic administration at 10mg/kg/week in a rodent study (Adelusi & Salako, 1982).

To successfully reposition CQ as an antiviral prophylactic against a respiratory virus such as COVID-19, we need to formulate an optimal dosing regimen which can achieve relevant viral inhibition in respiratory tissues with a margin of safety. Fortunately, since CQ has long been in use, we have extensive PK and toxicology data on the drug, including for children (Karunajeewa et al., 2008), in pregnancy (Lee et al., 2008), for short term prophylaxis against malaria as well as for long-term administration in autoimmune disease (Wollheim et al., 1978).

8

2.4 CQ COVID-19 prophylaxis – dose regimens

Current clinical dosing recommendations for CQ depends on indication. For malaria, the World Health Organization currently recommends the adult dose of 500 mg (base) weekly for prophylaxis, and 25mg/kg over 3 days for treatment for acute attack in uncomplicated cases (World Health Organization, 1995). In autoimmune diseases, the generally advocated dose is 250-500mg daily for rheumatoid arthritis (Popert et al., 1961) and 250 mg per day in SLE (Meinão et al., 1996).

Dose finding for a repurposed drug should be guided by effective drug levels against the target condition, as well as informed by dose ranges and known toxicities applied and reported from the drug's existing approved or indicated usage.

Dosage can also be guided by animal models as CQ PK in mice are similar to those reported for humans (Madrid et al., 2013) and rodent studies can also provide useful guidance for effective dosing in higher animals.

Established safe clinical application of CQ ranges from dosing of 500mg weekly in malaria prophylaxis to 500mg daily or more for acute malaria or chronic autoimmune conditions (Ducharme & Farinotti, 1996). These same dose range seems adequate to exert antiviral effects on hCoVs such as SARS-CoV (Vincent et al., 2005) and COVID-19 based on *in vitro* results (2.2 above).

The weekly CQ dose of 500mg for malaria prophylaxis yields only 0.9-1.3 μM in whole blood the day after treatment and troughs at 0.4-0.5 μM prior to the next dose (Rombo et al., 1987), which is below the EC_{50} for inhibition of COVID-19 and thus not optimal for COVID-19 prevention.

However, the low end of the dose range of CQ used for the treatment of rheumatoid arthritis (3.6 mg/kg or 250mg a day) generated plasma CQ concentrations of 1–1.6 μM (Wollheim et al., 1978), which would be in range of the EC_{50} for

COVID-19 inhibition (Wang et al., 2020). Separately, a higher but shorter dose of CQ for acute malaria at 8mg/kg/day for 3 days achieves a serum concentration of 9 μM (Marques et al., 2014) which is above the EC_{90} value of 6.90 μM against COVID-19 and can be adopted for post-exposure prophylaxis.

Based on the above analysis and synthesis, we recommend two prophylactic schedules for CQ antiviral against COVID-19:

1) CQ 8mg/kg/day for 3 days in post-exposure but asymptomatic cases, ideally to be taken within hours after known viral exposure based on *in vitro* data that CQ maybe significantly effective even 5 hours after virus adsorption and infection (Vincent et al., 2005).

2) CQ 500mg a day as chronic prophylaxis for people in outbreak locales or endemic areas with a high risk of exposure, to reduce to 250mg a day after 30 days to continue until the threat of infection is abated.

The higher initial dose of 500mg for chronic prophylaxis is based on achievable serum levels in the same range (Wollheim et al., 1978) of the EC_{50} and EC_{90} range of 1.13-6.90 μM against the virus (Wang et al., 2020) and we expect even higher tissue concentrations than in the serum so the dose is most likely adequate. The reduced dose of 250mg after 30 days of treatment is justified based on large increased and cumulative concentration in lung and other organ tissues after repeated dosing over time (Adelusi & Salako, 1982), as well as a concern for long-term toxicity after prolonged use (2.5 below).

2.5 CQ toxicity

CQ is generally considered safe and well tolerated with its side-effects well delineated. For relevance, we limit our review only to side-effects and potential

toxicities related to the two dose regimens proposed above for COVID-19 prophylaxis.

Our first proposed regimen is CQ 8mg/kg/day for 3 days in post-exposure but asymptomatic cases is similar to the treatment dose for acute malaria attack. The potential side-effects for this short duration regimen include nausea, anorexia, abdominal pain, vomiting, dizziness, headache, blurry vision and pruritus (Salako, 1984). A small phase 1 trial found these side-effects to be dose-related and generally under 15% except for headache which is the most common side-effect at 21% with doses comparable or slightly above this regimen's (Mzayek et al., 2007). These side-effects are usually mild, transient and can be minimized by taking CQ with food.

Our second proposed regimen is a prophylactic dose for those at high risk of acquiring the infection and is at 250-500mg daily for the duration of susceptibility, which could last months but is unlikely to go on for years. This dosing schedule is consistent with dosages used in autoimmune disorders. A relevant review on CQ toxicities related to chronic use at 250-500mg daily for SLE included 95 articles between 1982-2007 and confirms the general clinical experience that toxicity is infrequent, mild and usually reversible (Ruiz-Irastorza et al., 2010). Besides the above mentioned minor side-effects, chronic administration of CQ leads to tissue accumulation and pose a unique and rare set of toxicities including retinal, cardiac, ocular, neurologic. Retinal toxicity is a particularly serious concern in chronic use because of its debility. According to one report, the incidence of toxic ocular effects in less than 1% of adults treated such as CQ at 4mg/kg/day (approximately 250mg daily) for 5 years or less, but increases with duration of treatment (Marmor et al., 2016). Since we carefully chose a prolonged prophylaxis dose of 250mg per day after 30 days, and our proposed prophylactic use is intended for months and not years, the

11

ocular toxicity is not as relevant a concern, but a patient placed on CQ antiviral prophylaxis should be informed by the doctor on ocular adverse effects and receive regular monitoring (Wiacek et al., 2017).

As for teratogenicity in pregnancy, CQ can pass through the placenta, but the use of these drugs during pregnancy does not appear to risk harm to the fetus (Rainsford et al., 2015) , although harm cannot be excluded as there are a lack of studies.

In sum, decades long experience with the acute and chronic use up to years of various doses of CQ show a low incidence of adverse effects. The main concern in long-term administration is retinopathy and other tissue toxicities associated with drug accumulation, which we do not expect in prophylaxis against COVID-19 as the pandemic and hence the need of CQ prophylaxis is generally forecast to last for months and not years.

3. Discussion

Current inadequacies in containing the COVID-19 pandemic is evident by daily escalating numbers of infected cases and ever increasing territories succumbing to the virus, despite public containment efforts and personal preventative measures of citizens world-wide. As infections soar, healthcare systems will be taxed to the brink and fear and panic escalates. Pharmaceutical efforts involve rapid development of effective vaccines as well as discovery of novel therapeutics against the virus, but these efforts are costly and take time (DiMasi et al., 2016). Drug repositioning where existing drugs in the market with established safety profiles are redeployed for a new indication can lead to less costly and faster approval and deployment (Mullard, 2012),

and this approach should be especially considered when the urgent and timely need for effective therapeutics is needed as in the current case of COVID-19.

3.1 Antiviral prophylaxis in viral epidemics

Four major pharmacological prophylaxis to prevent and protect populations during a viral pandemic include vaccination (Zepp, 2016), passive neutralizing antibodies (Casadevall & Pirofski, 2015), convalescent plasma (Marano et al., 2016) and small molecule drugs (Madrid et al., 2013). There is active research on vaccine development as well as use of neutralizing antibodies and convalescent plasma for COVID-19 but currently no agent is ready to enter the clinic or near mass deployment for prevention (Li & De Clercq, 2020). Meanwhile, the current emphasis on antiviral drugs leans towards treatment rather than prevention.

Small molecule drugs as therapeutics against novel viruses has the advantage of stability and convenience of oral administration. Two development paths could be *de novo* synthesis of inhibitors targeting unique viral proteins involved in its infection process or screening for potential drug candidates in existing drug databases (Madrid et al., 2013). These approach has been deployed for other hCoVs with pandemic potential such as SARS-CoV (De Clercq, 2006) and Mers-CoV (Liang et al., 2018) and is underway for COVID-19, but the research and experience of deployment of these prophylactic measures is more established for the influenza virus.

Conceptually, massive antiviral prophylaxis might be effective in containing a viral pandemic as in the use of neuraminidase inhibitors against influenza (Saunders-Hastings et al., 2016). A Cochrane Collaboration review found that prophylactic use of antiviral neuraminidase inhibitors reduce the risk of developing influenza (Jefferson et al., 2014). Multiple randomized studies further demonstrated the utility

of neuraminidase inhibitors irrespective or pre- or post-exposure use in the rapid containment of influenza, offering 67-89% protection in individuals and households (Jefferson et al., 2014).

3.2 Drug repositioning against viruses and COVID-19

Given clinical experience of use and the fact that human safety studies have already been conducted, repositioned drugs offers many advantages as a path of least resistance for large-scale public deployment, especially in the midst of a rapidly advancing viral pandemic. Drug development risk, time, and cost are dramatically reduced because the drug candidates would have established safety and PK profiles, while chemical optimization, toxicology, bulk manufacturing, as well as formulation development have already been addressed (Strittmatter, 2014).

There is a long history of drug repositioning for viral diseases and there are currently around two dozen drugs and drug combination candidates for this purpose, targeting Zika, hCoVs, Influenza , Herpes, Norovirus, Rotavirus, and EBOV, some of which are already in phase 2/3 trials (Mercorelli et al., 2018). Specifically, strong cases have already been made previously to reposition existing drugs including CQ and its hydroxyl derivative hydroxychloroquine against hCoVs such as SARS-CoV (de Wilde et al., 2011) and MERS-CoV(De Wilde et al., 2014) and CQ repositioned as therapy for COVID-19 pneumonia is currently in multiple clinical trials in China (Gao et al., 2020) .

3.3 Repositioning CQ as ideal COVID-19 prophylactic

CQ has been called upon as a therapeutic agent against the hCoVs: MERS-CoV, SARS-CoV and now also COVID-19, but the emphasis has been on treatment for

symptomatic cases. On Feb 2020, based on encouraging preliminary findings from ongoing clinical trials in China, a government sponsored conference accepted the findings on CQ's potent activity against COVID-19 without considering is preventative potential and the drug has now been recommended for inclusion in the next version of the Guidelines for the Prevention, Diagnosis, and Treatment of Pneumonia Caused by COVID-19 issued by China's National Health Commission (Gao et al., 2020). Indeed, one of the latest official provincial Chinese government directives which came after reports of clinical efficacy of CQ specifically limits usage to confirmed cases of symptomatic adults between 18-65, and warns against prophylactic use, which we believe is a policy that maybe unjustifiably conservative (Multicenter Collaboration Group of Dept of Science and Technology and Health Commission of Guangdong Province for Chloroquine, 2020).

The key issues for developing antiviral drugs for prophylaxis in epidemics are 1) if the agent might work (scientific plausibility); 2) if the agent can be given at the right time to work (PK and PD); 3) if benefits outweigh cost and toxicities; and 4) if the agent could be readily available to a large population. CQ fulfils all the above criteria as a potential antiviral for prophylaxis against COVID-19. With respect to the above, we have reviewed here the *in vitro* data for CQ against COVID-19 as well as preliminary human trial data of its successful use in treatment of symptomatic patients. We have reviewed CQ's PK confirming its rapid absorption and ability to achieve antiviral concentrations in the body potentially at much greater concentration in relevant human tissue such as the lung. Furthermore, CQ's long history of extensive use, outstanding toxicity profile, as well as its international availability as a generic with numerous manufacturers worldwide including but not limited to Aventis, Alpharma, Bayer, Beltapharm, Cipla, Ecobi, Glaxosmithkline, Sanofi, Intas and Ipca,

as well a very low wholesale cost of under $0.10 per course of malaria treatment in the developing world (Arrow, G et al., 2004) all mark the drug as an ideal prophylactic in the COVID-19 pandemic.

3.4 Limitations, obstacles and further research

While there is convincing laboratory data that CQ inhibits COVID-19 at clinically relevant dosages and recent preliminary clinical data that CQ is efficacious in the treatment of clinical COVID-19 pneumonia, we lack an animal model to test (Broodman, 2020) and clinical data to confirm CQ as an effective antiviral prophylactic against COVID-19.

For example, studies have reported inhibitory effects of CQ against viruses such as influenza (Eng et al., 2006) and that treatment enhanced survival but was not effective as a prophylactic in rodents (Yan et al., 2013), and a clinical trial did not demonstrate effectiveness in prevention (Paton et al., 2011). Where CQ has also demonstrated impressive inhibition of CHIKV in a dose dependent manner (Sourisseau et al., 2007), a trial in the French Reunion Island during a CHIKV outbreak also did not show benefit (De Lamballerie et al., 2008). Thus it is possible that despite strong in vitro data supported by therapeutic efficacy in an ongoing Chinese clinical trial, CQ may yet prove to be ineffective as a prophylactic.

Proof of efficacy not withstanding, we see other majors obstacles in CQ for prophylaxis even if nations and governments rapidly adopt this as a public health measure with mass prescription and distribution of CQ. Bill Gates in an article on responding to COVID-19 in the New England Journal of Medicine this year summarized the challenges as technical, diplomatic and budgetary (Gates, 2020).

Despite such challenges, what is the alternative? In surveying the few antiviral prophylactic candidates in development or under consideration (Zhang & Liu, 2020) , CQ may currently be the best if not the only choice for rapid public deployment given its potential efficacy, safety record, existing manufacturing capacity, and low costs.

From the research angle, there is urgent need for private and public funding into basic science research on CQ's mode of action on COVID-19. Finding and testing an animal model for prophylactic efficacy, with further PD studies on important issues such as actual tissue concentration of the drug over repeat dosing would guide and help refine regimen design. Finally, the search, development and validation of related compounds or derivatives such as hydroxychloroquine that may be more efficacious and/or less toxic, clinical trials of CQ by itself and in combination with other potentially synergistic antivirals all deserve urgent and concerted attention.

4. Conclusion

CQ has very significant advantages as a lead candidate for antiviral prophylaxis against the current COVID-19 pandemic where no current vaccine or antiviral prophylaxis is in place. Its demonstrated mechanisms of action of preventing viral entry and fusion, evidence of in vitro efficacy at clinically achievable doses, high tissue concentration as well as preliminary clinical evidence of efficacy as treatment all support its promising preventative role. Its safety record and low cost at doses we propose imply a high potential benefit to risk and benefit to cost ratio when used for prophylaxis. We urge relevant agencies to consider initiating trials as well as prepare for direct mass deployment of a CQ based COVID-19 preventative program without undue delay.

References

Adelusi, S. A., & Salako, L. A. (1982). Tissue and blood concentrations of chloroquine following chronic administration in the rat. *Journal of Pharmacy and Pharmacology*, *34*(11), 733–735. https://doi.org/10.1111/j.2042-7158.1982.tb06211.x

Al-Bari, M. A. A. (2015). Chloroquine analogues in drug discovery: new directions of uses, mechanisms of actions and toxic manifestations from malaria to multifarious diseases. *Journal of Antimicrobial Chemotherapy*. https://doi.org/10.1093/jac/dkv018

Al-Bari, Md. Abdul Alim. (2017). Targeting endosomal acidification by chloroquine analogs as a promising strategy for the treatment of emerging viral diseases. *Pharmacology Research & Perspectives*, *5*(1), e00293. https://doi.org/10.1002/prp2.293

Arrow KJ, Panosian C, G. H. (Ed.). (2004). *Saving lives, buying time: economics of malaria drugs in an age of resistance*. National Academies Press. https://doi.org/10.17226/11017

Broodman E (STAT). (2020). *From ferrets to mice and marmosets, labs scramble to find right animals for coronavirus studies*. https://www.statnews.com/2020/03/05/coronavirus-labs-scramble-to-find-right-animals-for-covid-19-studies/

Casadevall, A., & Pirofski, L.-A. (2015). The Ebola epidemic crystallizes the potential of passive antibody therapy for infectious diseases. *PLoS Pathogens*, *11*(4), e1004717. https://doi.org/10.1371/journal.ppat.1004717

Coatney, G. R. (1963). Pitfalls in a Discovery: The Chronicle of Chloroquine. *The American Journal of Tropical Medicine and Hygiene*, *12*(2), 121–128. https://doi.org/10.4269/ajtmh.1963.12.121

De Clercq, E. (2006). Potential antivirals and antiviral strategies against SARS coronavirus infections. *Expert Review of Anti-Infective Therapy*, *4*(2), 291–302. https://doi.org/10.1586/14787210.4.2.291

De Lamballerie, X., Boisson, V., Reynier, J. C., Enault, S., Charrel, R. N., Flahault, A., Roques, P., & Grand, R. Le. (2008). On chikungunya acute infection and chloroquine treatment. *Vector-Borne and Zoonotic Diseases*. https://doi.org/10.1089/vbz.2008.0049

De Wilde, A. H., Jochmans, D., Posthuma, C. C., Zevenhoven-Dobbe, J. C., Van Nieuwkoop, S., Bestebroer, T. M., Van Den Hoogen, B. G., Neyts, J., & Snijder, E. J. (2014). Screening of an FDA-approved compound library identifies four small-molecule inhibitors of Middle East respiratory syndrome coronavirus

replication in cell culture. *Antimicrobial Agents and Chemotherapy*, *58*(8), 4875–4884. https://doi.org/10.1128/AAC.03011-14

de Wilde, A. H., Zevenhoven-Dobbe, J. C., van der Meer, Y., Thiel, V., Narayanan, K., Makino, S., Snijder, E. J., & van Hemert, M. J. (2011). Cyclosporin A inhibits the replication of diverse coronaviruses. *Journal of General Virology*. https://doi.org/10.1099/vir.0.034983-0

Desai, M., Field, N., Grant, R., & McCormack, S. (2017). Recent advances in pre-exposure prophylaxis for HIV. *BMJ (Clinical Research Ed.)*, *359*, j5011. https://doi.org/10.1136/bmj.j5011

DiMasi, J. A., Grabowski, H. G., & Hansen, R. W. (2016). Innovation in the pharmaceutical industry: New estimates of R&D costs. *Journal of Health Economics*. https://doi.org/10.1016/j.jhealeco.2016.01.012

Ducharme, J., & Farinotti, R. (1996). Clinical pharmacokinetics and metabolism of chloroquine. Focus on recent advancements. *Clinical Pharmacokinetics*, *31*(4), 257–274. https://doi.org/10.2165/00003088-199631040-00003

Eng, E. O., Chew, J. S. W., Jin, P. L., & Chua, R. C. S. (2006). In vitro inhibition of human influenza A virus replication by chloroquine. *Virology Journal*. https://doi.org/10.1186/1743-422X-3-39

Farias, K. J. S., Machado, P. R. L., Muniz, J. A. P. C., Imbeloni, A. A., & da Fonseca, B. A. L. (2015). Antiviral Activity of Chloroquine Against Dengue Virus Type 2 Replication in Aotus Monkeys. *Viral Immunology*, *28*(3), 161–169. https://doi.org/10.1089/vim.2014.0090

Gao, J., Tian, Z., & Yang, X. (2020). Breakthrough: Chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. *Bioscience Trends*. https://doi.org/10.5582/bst.2020.01047

Gates, B. (2020). Responding to Covid-19 - A Once-in-a-Century Pandemic? *The New England Journal of Medicine*. https://doi.org/10.1056/NEJMp2003762

Hoffmann, M., Kleine-Weber, H., Schroeder, S., Krüger, N., Herrler, T., Erichsen, S., Schiergens, T. S., Herrler, G., Wu, N.-H., Nitsche, A., Müller, M. A., Drosten, C., & Pöhlmann, S. (2020). SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell*. https://doi.org/10.1016/j.cell.2020.02.052

Hussein, I. T. M., Brooks, J., & Bowlin, T. L. (2020). The discovery and development of filociclovir for the prevention and treatment of human cytomegalovirus-related disease. *Antiviral Research*, 104710. https://doi.org/10.1016/j.antiviral.2020.104710

Jefferson, T., Jones, M. A., Doshi, P., Del Mar, C. B., Hama, R., Thompson, M. J., Spencer, E. A., Onakpoya, I., Mahtani, K. R., Nunan, D., Howick, J., & Heneghan, C. J. (2014). Neuraminidase inhibitors for preventing and treating influenza in healthy adults and children. *The Cochrane Database of Systematic Reviews*, *4*, CD008965. https://doi.org/10.1002/14651858.CD008965.pub4

Karunajeewa, H. A., Ilett, K. F., Mueller, I., Siba, P., Law, I., Page-Sharp, M., Lin, E., Lammey, J., Batty, K. T., & Davis, T. M. E. (2008). Pharmacokinetics and efficacy of piperaquine and chloroquine in melanesian children with uncomplicated malaria. *Antimicrobial Agents and Chemotherapy*. https://doi.org/10.1128/AAC.00555-07

Keyaerts, E., Li, S., Vijgen, L., Rysman, E., Verbeeck, J., Van Ranst, M., & Maes, P. (2009). Antiviral Activity of Chloroquine against Human Coronavirus OC43 Infection in Newborn Mice. *Antimicrobial Agents and Chemotherapy*, *53*(8), 3416–3421. https://doi.org/10.1128/AAC.01509-08

Keyaerts, Els, Vijgen, L., Maes, P., Neyts, J., & Van Ranst, M. (2004). In vitro inhibition of severe acute respiratory syndrome coronavirus by chloroquine. *Biochemical and Biophysical Research Communications*, *323*(1), 264–268. https://doi.org/10.1016/j.bbrc.2004.08.085

Kono, M., Tatsumi, K., Imai, A. M., Saito, K., Kuriyama, T., & Shirasawa, H. (2008). Inhibition of human coronavirus 229E infection in human epithelial lung cells (L132) by chloroquine: involvement of p38 MAPK and ERK. *Antiviral Research*, *77*(2), 150–152. https://doi.org/10.1016/j.antiviral.2007.10.011

Kouroumalis, E. A., & Koskinas, J. (1986). Treatment of chronic active hepatitis B (CAH B) with chloroquine: A preliminary report. *Annals of the Academy of Medicine Singapore*.

Krishna, S., & White, N. J. (1996). Pharmacokinetics of quinine, chloroquine and amodiaquine. Clinical implications. *Clinical Pharmacokinetics*, *30*(4), 263–299. https://doi.org/10.2165/00003088-199630040-00002

Kuchler, H., Cookson, C., & Neville, S. (2020, March 6). The $2bn race to find a vaccine. *Financial Times*, 7. https://www.ft.com/content/e0ecc6b6-5d43-11ea-b0ab-339c2307bcd4

Lee, S. J., McGready, R., Fernandez, C., Stepniewska, K., Paw, M. K., Viladpai-nguen, S. J., Thwai, K. L., Villegas, L., Singhasivanon, P., Greenwood, B. M., White, N. J., & Nosten, F. (2008). Chloroquine pharmacokinetics in pregnant and nonpregnant women with vivax malaria. *European Journal of Clinical Pharmacology*, *64*(10), 987–992. https://doi.org/10.1007/s00228-008-0500-z

Li, C., Zhu, X., Ji, X., Quanquin, N., Deng, Y.-Q., Tian, M., Aliyari, R., Zuo, X., Yuan, L., Afridi, S. K., Li, X.-F., Jung, J. U., Nielsen-Saines, K., Qin, F. X.-F., Qin, C.-F., Xu, Z., & Cheng, G. (2017). Chloroquine, a FDA-approved Drug, Prevents Zika Virus Infection and its Associated Congenital Microcephaly in Mice. *EBioMedicine*, *24*, 189–194. https://doi.org/10.1016/j.ebiom.2017.09.034

Li, G., & De Clercq, E. (2020). Therapeutic options for the 2019 novel coronavirus (2019-nCoV). *Nature Reviews Drug Discovery*. https://doi.org/10.1038/d41573-020-00016-0

Liang, R., Wang, L., Zhang, N., Deng, X., Su, M., Su, Y., Hu, L., He, C., Ying, T., Jiang, S., & Yu, F. (2018). Development of Small-Molecule MERS-CoV Inhibitors. *Viruses*, *10*(12). https://doi.org/10.3390/v10120721

Long, J., Wright, E., Molesti, E., Temperton, N., & Barclay, W. (2015). Antiviral therapies against Ebola and other emerging viral diseases using existing medicines that block virus entry. *F1000Research*, *4*, 30. https://doi.org/10.12688/f1000research.6085.2

Madrid, P. B., Chopra, S., Manger, I. D., Gilfillan, L., Keepers, T. R., Shurtleff, A. C., Green, C. E., Iyer, L. V., Dilks, H. H., Davey, R. A., Kolokoltsov, A. A., Carrion, R., Patterson, J. L., Bavari, S., Panchal, R. G., Warren, T. K., Wells, J. B., Moos, W. H., Burke, R. L., & Tanga, M. J. (2013). A Systematic Screen of FDA-Approved Drugs for Inhibitors of Biological Threat Agents. *PLoS ONE*, *8*(4), e60579. https://doi.org/10.1371/journal.pone.0060579

Manic, G., Obrist, F., Kroemer, G., Vitale, I., & Galluzzi, L. (2014). Chloroquine and hydroxychloroquine for cancer therapy. In *Molecular and Cellular Oncology*. https://doi.org/10.4161/mco.29911

Marano, G., Vaglio, S., Pupella, S., Facco, G., Catalano, L., Liumbruno, G. M., & Grazzini, G. (2016). Convalescent plasma: New evidence for an old therapeutic tool? In *Blood Transfusion*. https://doi.org/10.2450/2015.0131-15

Marmor, M. F., Kellner, U., Lai, T. Y. Y., Melles, R. B., Mieler, W. F., & Lum, F. (2016). Recommendations on Screening for Chloroquine and Hydroxychloroquine Retinopathy (2016 Revision). *Ophthalmology*. https://doi.org/10.1016/j.ophtha.2016.01.058

Meinão, I. M., Sato, E. I., Andrade, L. E., Ferraz, M. B., & Atra, E. (1996). Controlled trial with chloroquine diphosphate in systemic lupus erythematosus. *Lupus*, *5*(3), 237–241. https://doi.org/10.1177/096120339600500313

Mercorelli, B., Palù, G., & Loregian, A. (2018). Drug Repurposing for Viral Infectious Diseases: How Far Are We? *Trends in Microbiology*, *26*(10), 865–876. https://doi.org/10.1016/j.tim.2018.04.004

Mullard, A. (2012). Drug repurposing programmes get lift off. In *Nature Reviews Drug Discovery* (Vol. 11, Issue 7, pp. 505–506). https://doi.org/10.1038/nrd3776

multicenter collaboration group of Department of Science and Technology of Guangdong Province and Health Commission of Guangdong Province for chloroquine in the treatment of novel coronavirus pneumonia. (2020). [Expert consensus on chloroquine phosphate for the treatment of novel coronavirus pneumonia]. *Zhonghua Jie He He Hu Xi Za Zhi = Zhonghua Jiehe He Huxi Zazhi = Chinese Journal of Tuberculosis and Respiratory Diseases*, *43*, E019. https://doi.org/10.3760/cma.j.issn.1001-0939.2020.0019

Mzayek, F., Deng, H., Mather, F. J., Wasilevich, E. C., Liu, H., Hadi, C. M., Chansolme, D. H., Murphy, H. A., Melek, B. H., Tenaglia, A. N., Mushatt, D. M., Dreisbach, A. W., Lertora, J. J. L., & Krogstad, D. J. (2007). Randomized dose-ranging controlled trial of AQ-13, a candidate antimalarial, and chloroquine in healthy volunteers. *PLoS Clinical Trials*, *2*(1), e6. https://doi.org/10.1371/journal.pctr.0020006

Nap, R. E., Andriessen, M. P. H. M., Meessen, N. E. L., & van der Werf, T. S. (2007). Pandemic influenza and hospital resources. *Emerging Infectious Diseases*, *13*(11), 1714–1719. https://doi.org/10.3201/eid1311.070103

Oxford, J. S. (2007). Antivirals for the treatment and prevention of epidemic and pandemic influenza. *Influenza and Other Respiratory Viruses*, *1*(1), 27–34. https://doi.org/10.1111/j.1750-2659.2006.00006.x

Paton, N. I., Lee, L., Xu, Y., Ooi, E. E., Cheung, Y. B., Archuleta, S., Wong, G., & Smith, A. W. (2011). Chloroquine for influenza prevention: A randomised, double-blind, placebo controlled trial. *The Lancet Infectious Diseases*. https://doi.org/10.1016/S1473-3099(11)70065-2

Peters, W. (1971). Malaria. Chemoprophylaxis and chemotherapy. *British Medical Journal*, *2*(5753), 95–98. https://doi.org/10.1136/bmj.2.5753.95

Plantone, D., & Koudriavtseva, T. (2018). Current and Future Use of Chloroquine and Hydroxychloroquine in Infectious, Immune, Neoplastic, and Neurological Diseases: A Mini-Review. In *Clinical Drug Investigation*. https://doi.org/10.1007/s40261-018-0656-y

POPERT, A. J., MEIJERS, K. A., SHARP, J., & BIER, F. (1961). Chloroquine diphosphate in rheumatoid arthritis. A controlled trial. *Annals of the Rheumatic Diseases*, *20*, 18–35. https://doi.org/10.1136/ard.20.1.18

Rainsford, K. D., Parke, A. L., Clifford-Rashotte, M., & Kean, W. F. (2015). Therapy and pharmacological properties of hydroxychloroquine and chloroquine in treatment of systemic lupus erythematosus, rheumatoid arthritis and related diseases. In *Inflammopharmacology*. https://doi.org/10.1007/s10787-015-0239-y

Randolph, V. B., Winkler, G., & Stollar, V. (1990). Acidotropic amines inhibit proteolytic processing of flavivirus prM protein. *Virology*, *174*(2), 450–458. https://doi.org/10.1016/0042-6822(90)90099-d

Rezaee, F., Linfield, D. T., Harford, T. J., & Piedimonte, G. (2017). Ongoing developments in RSV prophylaxis: a clinician's analysis. *Current Opinion in Virology*, *24*, 70–78. https://doi.org/10.1016/j.coviro.2017.03.015

Rolain, J. M., Colson, P., & Raoult, D. (2007). Recycling of chloroquine and its hydroxyl analogue to face bacterial, fungal and viral infections in the 21st century. In *International Journal of Antimicrobial Agents*. https://doi.org/10.1016/j.ijantimicag.2007.05.015

Rombo, L., Bergqvist, Y., & Hellgren, U. (1987). Chloroquine and desethylchloroquine concentrations during regular long-term malaria prophylaxis. *Bulletin of the World Health Organization*.

Ruiz-Irastorza, G., Ramos-Casals, M., Brito-Zeron, P., & Khamashta, M. A. (2010). Clinical efficacy and side effects of antimalarials in systemic lupus erythematosus: A systematic review. In *Annals of the Rheumatic Diseases*. https://doi.org/10.1136/ard.2008.101766

Salako, L. A. (1984). Toxicity and side-effects of antimalarials in Africa: A critical review. *Bulletin of the World Health Organization*.

Saunders-Hastings, P., Reisman, J., & Krewski, D. (2016). Assessing the state of knowledge regarding the effectiveness of interventions to contain pandemic influenza transmission: A systematic review and narrative synthesis. *PLoS ONE*, *11*(12), 1–17. https://doi.org/10.1371/journal.pone.0168262

Savarino, A., Boelaert, J. R., Cassone, A., Majori, G., & Cauda, R. (2003). Effects of chloroquine on viral infections: An old drug against today's diseases? In *Lancet Infectious Diseases*. https://doi.org/10.1016/S1473-3099(03)00806-5

Shimizu, Y., Yamamoto, S., Homma, M., & Ishida, N. (1972). Effect of chloroquine on the growth of animal viruses. *Archiv Fur Die Gesamte Virusforschung*, *36*(1), 93–104. https://doi.org/10.1007/bf01250299

Sourisseau, M., Schilte, C., Casartelli, N., Trouillet, C., Guivel-Benhassine, F., Rudnicka, D., Sol-Foulon, N., Le Roux, K., Prevost, M. C., Fsihi, H., Frenkiel, M. P., Blanchet, F., Afonso, P. V., Ceccaldi, P. E., Ozden, S., Gessain, A., Schuffenecker, I., Verhasselt, B., Zamborlini, A., … Schwartz, O. (2007). Characterization of reemerging chikungunya virus. *PLoS Pathogens*. https://doi.org/10.1371/journal.ppat.0030089

Sperber, K., Louie, M., Kraus, T., Proner, J., Sapira, E., Lin, S., Stecher, V., & Mayer, L. (1995). Hydroxychloroquine treatment of patients with human immunodeficiency virus type 1. *Clinical Therapeutics*, *17*(4), 622–636. https://doi.org/10.1016/0149-2918(95)80039-5

Strittmatter, S. M. (2014). Overcoming Drug Development Bottlenecks With Repurposing: Old drugs learn new tricks. *Nature Medicine*, *20*(6), 590–591. https://doi.org/10.1038/nm.3595

V., T., N.N., M., T.P., V., S.J., L., J., F., B., W., & H.T., T. (2010). A randomized controlled trial of chloroquine for the treatment of dengue in vietnamese adults. *PLoS Neglected Tropical Diseases*.

Vincent, M. J., Bergeron, E., Benjannet, S., Erickson, B. R., Rollin, P. E., Ksiazek, T. G., Seidah, N. G., & Nichol, S. T. (2005). Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virology Journal*, *2*. https://doi.org/10.1186/1743-422X-2-69

Wang, M., Cao, R., Zhang, L., Yang, X., Liu, J., Xu, M., Shi, Z., Hu, Z., Zhong, W., & Xiao, G. (2020). Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Research*, *30*(3), 269–271. https://doi.org/10.1038/s41422-020-0282-0

White, N. J., Pukrittayakamee, S., Hien, T. T., Faiz, M. A., Mokuolu, O. A., & Dondorp, A. M. (2014). Malaria. *The Lancet*, *383*(9918), 723–735. https://doi.org/10.1016/S0140-6736(13)60024-0

Wiacek, M. P., Bobrowska-Snarska, D., Lubiński, W., Brzosko, M., & Modrzejewska, M. (2017). What is new in recommendations on ophthalmological screening in patients treated with chloroquine and hydroxychloroquine? Update and literature review. *Nigerian Journal of Clinical Practice*, *20*(8), 919–923. https://doi.org/10.4103/njcp.njcp_380_16

Wollheim, F. A., Hanson, A., & Laurell, C.-B. (1978). Chloroquine Treatment in Rheumatoid Arthritis. *Scandinavian Journal of Rheumatology*, 7(3), 171–176. https://doi.org/10.3109/03009747809095649

World Health Organization. (1995). WHO Model Prescribing Information: Drugs Used in Parasitic Diseases - Second Edition. In *WHO Model Prescribing Information: Drugs Used in Parasitic Diseases - Second Edition*.

Yan, Y., Zou, Z., Sun, Y., Li, X., Xu, K.-F., Wei, Y., Jin, N., & Jiang, C. (2013). Anti-malaria drug chloroquine is highly effective in treating avian influenza A H5N1 virus infection in an animal model. *Cell Research*, *23*(2), 300–302. https://doi.org/10.1038/cr.2012.165

Zepp, F. (2016). Principles of Vaccination. *Methods in Molecular Biology (Clifton, N.J.)*, *1403*, 57–84. https://doi.org/10.1007/978-1-4939-3387-7_3

Zhang, L., & Liu, Y. (2020). Potential Interventions for Novel Coronavirus in China: A Systemic Review. *Journal of Medical Virology*. https://doi.org/10.1002/jmv.25707

Zhu, N., Zhang, D., Wang, W., Li, X., Yang, B., Song, J., Zhao, X., Huang, B., Shi, W., Lu, R., Niu, P., Zhan, F., Ma, X., Wang, D., Xu, W., Wu, G., Gao, G. F., & Tan, W. (2020). A Novel Coronavirus from Patients with Pneumonia in China, 2019. *New England Journal of Medicine*, *382*(8), 727–733. https://doi.org/10.1056/NEJMoa2001017

26