Focused Review

COVID-19 Pandemic - A Narrative Review of the Potential Roles of Chloroquine and Hydroxychloroquine

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Background: Chloroquine (CQ) and hydroxychloroquine (HCQ) are old drugs used against malaria, rheumatism, inflammation in the joints, lupus, among others. These drugs showed positive results in preliminary scientific research for treatment of the severe acute respiratory syndrome coronavirus (SARS-CoV-2). Since the studies with CQ and HCQ are initial with small patient populations, it is not yet known whether there are adverse effects from the use of CQ and HCQ for patients infected with the coronavirus.

Objectives: The aim of this study was to evaluate the evidence regarding the efficacy and safety of CQ and HCQ used against viral infection caused by SARS-CoV-2.

Study Design: This is a narrative review of the traditional prescriptions of CQ and HCQ efficacy and adverse effects as well as their employment for coronavirus disease 2019 (COVID-19).

Setting: In vitro and clinical studies comparing the antiviral efficacy and adverse effect profile of CQ and HCQ against COVID-19 in adult patients were evaluated.

Methods: A systemic search of reviews, including in vitro and clinical trial studies in English focusing on CQ and HCQ effects and adverse effects against COVID-19 in the adult patient population from PubMed was performed. It included studies reporting chloroquine and hydroxychloroquine effects and adverse effects against COVID-19.

Results: A total of 42 articles published between 2004 and April 2020 were reviewed for therapeutic use of CQ and HCQ. Both these drugs showed a significant in vitro potential against coronavirus. Many studies for clinical use of CQ and HCQ showed that patients presented adverse reactions on high doses.

Limitations: Clinical studies have some methodology shortcomings, such as lack of information about the treatment and small number of experimental patients, leading to a misinterpretation of the data. Besides, there are few clinical studies with a limited sample size. Moreover, most of them did not present control groups, and some patients had died during these protocols.

Discussion: Despite both CQ and HCQ in vitro antiviral evidence, clinically, both drugs, either alone or combined with other medications, may increase the risk of cardiac arrhythmias, leading to cardiac arrest and sudden death. Besides, a lot of uncertainty still remains, such as starting administration period, dose prescribed, length of treatment, patients' condition, concomitant drug use, among others.

Conclusion: From the studies reviewed, it is not possible to state the precise efficacy and safety of CQ and HCQ use in the treatment of COVID-19 at any time in the course of the disease. Future studies are warranted.

Key words: SARS-CoV-2; COVID-19; chloroquine; hydroxychloroquine; pandemic; coronavirus; adverse effects; side effects; cardiac arrhythmias; ocular toxicity, retinopathy; pharmacokinetic.

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Recently discovered, an infection with the new severe acute respiratory syndrome coronavirus (SARS-CoV-2) has quickly spread worldwide. The outbreak of coronavirus disease 2019 (COVID-19) started in Hubei Province (Wuhan, China) in December of 2019 (1). Although genetically related to both SARS-CoV-1 and Middle East Respiratory Syndrome coronavirus (MERS-CoV), with a better genome sequence identity with SARS-CoV-1, the new SARS-CoV-2 presents higher infectiousness rates (2,3). Because of its fast dissemination both inside and outside China, COVID-19 was considered as a pandemic in March of 2020 by the World Health Organization (4).

The COVID-19 has a wide variability in clinical presentations, ranging from asymptomatic or mildly symptomatic to severe respiratory and systemic conditions resulting in death, especially in elderly patients and those with previous cardiovascular or respiratory comorbidities. In a study regarding the severity of the outbreak in China, WU and McGoogan (2020) (5) showed that from 72,314 registered cases, 81% were considered as mild cases, whereas 14% accounted for severe cases and 5% for critical cases. The age range affected was between 30 and 79 years old (87% of cases).

The transmission occurs mainly via respiratory droplet, although oral-fecal transmission has also been reported (6). The pathophysiology of COVID-19 can be divided into three main phases: 1) an early phase, characterized by both viral infiltration and replication, presenting lymphocytopenia; 2) a pulmonary phase, which is a progressive state of the disease, characterized by pulmonary damage, accompanied by respiratory compromising which can be detected by abnormal chest imaging; and 3) the inflammatory response overreaction, with an increase in proinflammatory markers production, known as cytokine storm syndrome (7).

In this context, drug repositioning appears as a possibility for new treatments against COVID-19. It is a new therapeutic use of an old drug, besides the original prescriptions. The COVID pandemic has raised several off-label and compassionate therapeutic possibilities for infected patients, most of them based on their in vitro antiviral activities and also their anti-inflammatory responses, considering COVID-19 pathophysiology. Among these drugs, the off-label chloroquine (CQ) and hydroxychloroquine (HCQ), drugs used in clinical medicine not only as antimalarial, but also for the treatment of autoimmune diseases, such as systemic lupus erythematosus and rheumatoid arthritis, have been used (8).

There are several mechanisms of action described for these antimalarial drugs, most being based on in vitro studies (9). Both drugs are bases that contain a large volume of distribution, accumulating on lysosomes and inhibiting endocytosis, phagocytosis and autophagy. In addition, these medications also inhibit signaling pathways that may reduce proinflammatory cytokines. They also interfere in the immune activation in different cellular levels, preventing the occurrence of many innate and adaptive immune processes.

The in-depth analysis of literature since the mid-1960s, CQ has shown to possess antiviral activity. In 1969, Inglot, Anna D. (10) compared the antiviral activity in vitro of several anti-inflammatory drugs. Further studies have proved that this antiviral action was probably due to the increase of the pH in the acid organelles, such as endosomes, phagosomes and Golgi apparatus (11).

The in vitro experiments performed by Wang, M., Cao, R., Zhang, L., et al (12) with CQ and HCQ were successful in treating the infection with the new coronavirus. Both drugs inhibit quinone reductase 2, interfering with sialic acid biosynthesis, crucial viruses' components used as receptors to enter human cells (13). Another described mode of action includes the glycosylation deficit of the virus cell surface receptors, compromising their binding to angiotensin-converting enzyme 2 (ACE2) receptors (14). However, due to the lack of in vivo efficacy, it is necessary Interventional studies to determine if these relationships are causal or merely associational.

The Infectious Diseases Society of America (IDSA) guideline (15) used for the management and treatment of the new coronavirus emphasizes the importance of additional randomized clinical studies, as well as the prospective results records, and it recommends the CQ and HCQ use in hospitalized patients in these clinical studies. The treatments with CQ and HCQ for COVID-19 are off-label (16).

However, the CQ or HCQ efficacy against COVID-19 is controversial and it has not been fully investigated. Thus, we performed a detailed pharmacological scientific review regarding the CQ and HCQ use as a treatment for SARS-CoV-2 and others coronavirus, exploring its pharmacokinetics and its adverse effects, as well as its risks and effectiveness.

Methods

Literature Search Strategy

An exhaustive search was performed on the

PubMed-Medline using the following keywords: COVID-19, Chloroquine and Hydroxychloroquine. We included all studies related to COVID-19, CQ and HCQ in vitro and clinical studies. Once the search was not restricted by publication date, all original papers regarding clinical, pharmacological and in vitro studies, as well as reviews published up to April 13th, 2020 were included. Fig. 1 shows the flowchart and each step performed in the selection process to recover relevant studies.

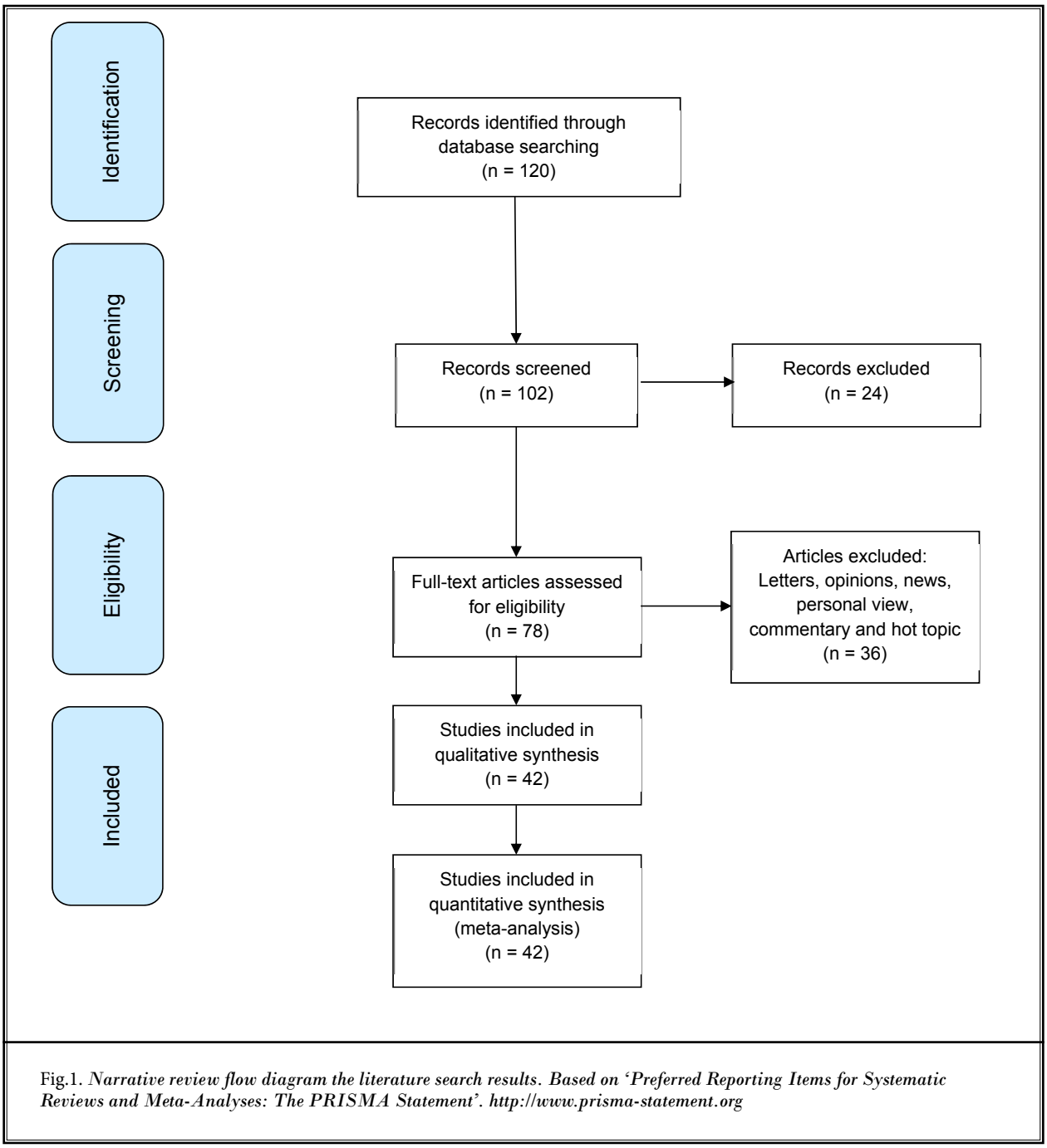

Fig.1. *Narrative review flow diagram the literature search results. Based on 'Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement'. http://www.prisma-statement.org*

Studies Selection: Inclusion and Exclusion Criteria

The initial literature search yielded more than 100 publications from PubMed. Only 78 articles fulfilled the inclusion criteria. These articles were then independently evaluated based on inclusion criteria by 5 investigators, and disagreements between the investigators were discussed and resolved. A total of 42 studies were included for the final analysis (Table 1). Considering the

Table 1. *Recent articles evaluating the potential roles of CQ and HCQ as treatments for COVID-19.*

Author/Date	Study Type	Method/Patients	Conclusion	Outcome
Ferrey et al., 2020 (17)	Case report	1 patient positive for COVID-19 with unusual symptoms.	A 56-year-old man with end-stage renal disease, who developed COVID-19 infection remained in critical condition being treated with HCQ and tocilizumab.	Not applicable
Spezzani, Piunno and Iselin, 2020 (18)	Case report	2 patients positive for COVID-19.	An immunocompromised woman (recent chemotherapy) and her presumably healthy husband (controlled arterial hypertension) were treated with darunavir/cobicistat and HCQ. The woman became negative for SARS-CoV-2 first. Her husband needed intensive care, however he was successful as well.	Not applicable
Inciardi et al., 2020 (19)	Case report	1 patient without previous cardiovascular disease and positive for SARS-CoV-2.	The cardiac involvement is a possible late phenomenon of the viral respiratory infection. Virus infection has been described as one of the most common infectious causes of myocarditis.	Not applicable
Bartiromo et al., 2020 (20)	Case report	1 kidney transplanted to a patient affected by Senior-Loken syndrome, positive for COVID-19.	Many therapies were applied during the treatment, including HCQ (200 mg bid) and after 9 days, the patient could be discharged from the hospital, and stayed at home isolation taking only corticosteroids.	Not applicable
Jun et al., 2020 (21)	Clinical study	30 patients randomized 1:1 to HCQ group and control group. HCQ dose 400mg/d per 5 days). Combination with conventional treatment (not related).	The results demonstrated that both HCQ and the control groups had an improvement to combat COVID-19, although larger sample size study is needed to investigate the effects of HCQ in COVID -19.	Favorable
Xu et al., 2020 (22)	Clinical study	62 patients with a history of traveling to Wuhan, positive for SARS-CoV-2 were observed for two weeks.	Patients from Zhejiang province had milder symptoms than patients from Wuhan.	Not applicable
Gautret et al., 2020 (23)	Clinical study	Non-randomized and not hidden study. Patients received isolated HCQ (600mg/daily) or in combination with azithromycin. Groups: HCQ and HCQ + azithromycin (n=20); control group (n=16). There is no specific treatment for control group. Lack of information.	A comparative analysis of the groups showed that on the sixth day, 14 of the 20 patients who received treatment with HCQ had negated the PCR against only 2 of the 16 patients in the control group. In the combined treatment group, 100% of the patients negated PCR on the fifth day of treatment. Three patients from the HCQ group were transferred to the intensive care unit, one died on day 3, one was lost to follow up and one stopped due to nausea. No control patients were lost to follow-up. Control group and intervention group treated in different locations.	Favorable
Perinel et al., 2020 (24)	Clinical Study	N=13 patients.	Only 61% of the patients that were treated with HCQ achieved the supposed minimum therapeutic level (1 mg/L). Also, HCQ was able to cause a severe adverse effect. More kinetic studies are still necessary to define the optimal dosing regimen for patients with COVID-19.	Not applicable
Huang et al., 2020 (25)	Clinical Study	The article demonstrates a comparative clinical trial between lopinavir/ ritonavir and CQ efficacy in COVID-19 patients (n=22).	Patients treated with CQ became negative for SARS-CoV-2, recovered better and regained their pulmonary function faster than the group who received lopinavir/ritonavir.	Favorable
Chen et al., 2020 (26)	Clinical Study	62 patients. HCQ treatment group (n=31) and control group (n=31).	The HCQ treatment in COVID-19 patients was efficient, since it was able to shorten the time to clinical recovery and promoted the improvement of pneumonia.	Favorable

Table 1 (cont.). *Recent articles evaluating the potential roles of CQ and HCQ as treatments for COVID-19.*

Author/Date	Study Type	Method/Patients	Conclusion	Outcome
Fantini et al., 2020 (27)	In silico	In silico study to assess the efficiency of CQ and HCQ as repositioned candidates for the treatment against the SARS-CoV-2 before their clinical evaluation.	A new type of ganglioside-binding domain (conserved among all clinical isolates) was shown and was able to improve the viral attachment and facilitate contact with the ACE2 receptor. The CQ and HCQ prevented the binding of the protein S to gangliosides.	Not applicable
Yao et al., 2020 (28)	In vitro	It used SARS-CoV-2 infected Vero-cells on PBPK models at 5 different doses.	HCQ was more potent than CQ. HCQ dose was 400mg twice a day, followed by a maintenance of 200mg twice a day for 4 days. CQ dose was 500 mg twice per 5 days.	Favorable
Keyaerts et al., 2004 (29)	In vitro	Cells infected with SARS-CoV were tested with isolated CQ-phosphate.	CQ inhibited SARS-CoV replication in Vero E6 cells on IC 50 of 8.8 μM.	Favorable
Vincent et al., 2005 (14)	In vitro	Vero E6 cells treated with CQ (isolated) before virus SARS-CoV infection.	CQ identified as an effective antiviral agent for SARS-CoV in cell culture. The drug was added prior to infection or after the establishment of infection. The antiviral effect suggests prophylactic and therapeutic advantages.	Favorable
Keyaerts et al., 2009 (30)	In Vitro and In vivo	CQ was added (10 μM) 24h prior to SARS-CoV infection and 3 to 5 hours after infection in HRT-18 cells. C57BL/6 pregnant mice were injected with CQ (different dilutions) and subsequently, puppies were inoculated with the HCoV-OC43.	CQ showed in vitro antiviral properties against HCoV-OC43 replication in HRT-18 cells and against SARS-CoV infection. The inhibition of HCoV-OC43 replication was more potent. An antiviral effect before and after the establishment of the infection is suggested.	Not applicable
Barnard et al., 2006 (31)	In vitro and In vivo	African green monkey kidney cells (Vero 76); female BALB/c mice.	The use of different formulations of CQ (monophosphate, diphosphate, and the CQ itself) in different assays to verify their capacity to inhibit the SARS-CoV replication in vitro and in vivo, showing that in vitro had better results than in vivo. These drugs are possible treatments for SARS-CoV-2 infection.	Favorable
Devaux et al., 2020 (32)	Review	Narrative review.	CQ was able to interfere in the ideal cleavage of the virus and in the viral communication with its target cell through the inhibition of kinases pathway. Moreover, it was able to interfere with the proteolytic processing of the viral protein M.	Favorable
Du and Chen, 2020 (33)	Review	Narrative review.	Some CQ studies were presented; however, the focus is especially in the use of favipiravir.	Not applicable
Gbinigie and Frir, 2020 (34)	Review	Narrative review	There is no sufficient data to determine if CQ and HCQ are both effective and safe for the treatment of COVID-19.	Unfavorable
Kakodkar, Kaka and Baig, 2020 (35)	Review	Narrative review.	There are some characteristics about the coronavirus itself and the possible treatments regarding its related disease. One of the discussed treatments includes the CQ usage and applicability.	Neutral
Shah et al., 2020 (36)	Review	Narrative review.	Effects of CQ and HCQ can show contradictory results depending on the study. However, it is early to recommend these drugs as prophylaxis for COVID-19, since no clinical and in vivo data were published.	Not applicable
Cortegiani et al., 2020 (37)	Review	Narrative review.	Six articles were analyzed and all of them showed effectiveness of CQ for treatment of COVID-19, highlighting the importance of monitoring patients, due its side effects.	Favorable
Lai et al., 2020 (38)	Review	Narrative review.	CQ is a promising drug in COVID-19. Although clinical trials are investigating the efficacy of several agents, so far there is no effective treatment for SARS-CoV-2.	Not clear
Kapoor et al., 2020 (39)	Review	Narrative review.	The use of HCQ and their cardiovascular risks are taken into consideration, since HCQ has been used globally for treatment and prophylaxis of COVID-19, based on in vitro or some clinical data. The National Task Force for COVID-19 constituted by Indian Council of Medical Research (ICMR) recommended HCQ for prophylactic use.	Neutral

Table 1 (cont.). *Recent articles evaluating the potential roles of CQ and HCQ as treatments for COVID-19.*

Author/Date	Study Type	Method/Patients	Conclusion	Outcome
Pereira, 2020 (40)	Review	Narrative review.	Main side effects of CQ/HCQ are related to retinopathy, neuromyopathy, and cardiomyopathy. Both are slowly excreted due to the long half-life. This leads to tissue bioaccumulation after chronic treatments, being necessary to monitor side effects after the treatment is finished.	Not clear
Haslak et al., 2020 (41)	Review	Narrative review.	COVID-19 seems to be rare or has a milder course in children and rheumatic diseases did not represent a risk factor for more severe disease courses. An explanation can be that antirheumatic drugs, like HCQ, may have a protective and therapeutic role in COVID-19.	Favorable
Singh et al., 2020 (42)	Review	Narrative review.	Describes the mechanism of action of CQ and HCQ and mentions positive studies in the treatment of COVID-19 by these drugs.	Favorable
Gupta, Agrawal and Ish, 2020 (43)	Review	Narrative review.	There is not enough evidence to use CQ for treatment of COVID-19. The use should be restricted to clinical trials with strict vigilance and follow up to further clarify its role.	Unfavorable
Rosa and Santos, 2020 (44)	Review	Narrative review.	Repositioning clinical trials may represent a strategy which would facilitate the discovery of new classes of medicines, decrease costs and take less time to reach the market, and there are existing pharmaceutical supply chains for formulation and distribution.	Favorable
Gupt and Misra, 2020 (45)	Review	Narrative review.	HCQ could be offered as an off-label treatment to patients with moderate to severe COVID-19 infection.	Favorable
Zhang et al., 2020 (46)	Review	Narrative review.	CQ and its derivatives inhibit viral replication in vitro. However, there are potential adverse drug reactions, such as cardiotoxicity and irreversible retinopathy.	Unfavorable
Dashraath, Jeslyn and Karen, 2020 (47)	Review	Narrative review.	There is a necessity for a higher dose of CQ in pregnancy due to significant lower plasma concentrations. However, these higher doses could cause systolic hypotension.	Unfavorable
Hussain, Bhowmik and do Vale, 2020 (48)	Review	Narrative review.	To date, there is no consensus regarding the appropriate treatment for patients with diabetes and SARS-CoV-2-positive, as well as patients with COVID-19 who develop glycemic decompensation.	Unfavorable
Zhou, Dai and Tong, 2020 (49)	Review	Narrative review.	HCQ exhibits an antiviral effect highly similar to that of CQ, and could serve as a better therapeutic approach, due to CQ severe side effects.	Favorable
Wenzhong and Hualan, 2020 (50)	Review	Narrative review.	CQ is shown to be capable of preventing orf1ab, ORF3a, and ORF10 to attack the heme protein and forming porphyrin, which could at some extent decrease the symptoms of respiratory distress.	Favorable
Plantone and Koudriavtseva, 2018 (51)	Review	Narrative review.	HCQ and CQ have anti-inflammatory, immunomodulating, anti-infective, antithrombotic, and metabolic effects, as well as antitumoral properties.	Favorable
Agrawal, Goel and Gupta, 2020 (52)	Review	Brief narrative review.	Some studies have already been done with HCQ, and there are benefits from the use of this drug, however, there is not yet enough studies proving HCQ safety and efficacy.	Unfavorable
Xie and Chen, 2020 (53)	Review	Narrative review.	CQ has shown a significant inhibition of SARS-CoV multiplication in vitro and in vivo, as well as efficacy and acceptable safety in patients infected with COVID-19 in clinical trials.	Neutral
Sanders, Monogue and Jodlowski, 2020 (54)	Review	Narrative review.	More studies are necessary to point out if CQ/HCQ are safe and effective to treat COVID-10, and to reach an optimal dose of CQ and HCQ.	Unfavorable
Lu, Chen and Chang, 2020 (55)	Review	Narrative review.	CQ and HCQ showed inhibitory activity against SARS-Cov-2 in vitro and promising results in clinical studies. However, more clinical trials are necessary to confirm their efficacy and safety.	Neutral
Juurlink, 2020 (56)	Review	Narrative review.	The use of either CQ or HCQ and azithromycin for treatment or prevention of SARS-CoV-2 infection is currently supported primarily by in vitro data and few studies involving humans, and there are several uncommon but potentially life-threatening adverse effects.	Unfavorable
Zhai et al., 2020 (57)	Review	Narrative review.	CQ/HCQ demonstrated optimistic results in vitro and in a clinical trial. However, it is still necessary to determine whether the efficacy of CQ and its analogs are dependent on variables of the disease.	Favorable

heterogeneity in methodology and treatment indications among studies published between 2004 and 2020, a narrative review of these studies was performed.

In the present review, there are different categories of articles, including case reports and clinical studies. In this concept, there are 26 reviews, 4 case reports, 1 in silico study, 2 articles describing both in vitro and in vivo studies, 3 articles describing only in vitro studies and 6 clinical studies. All letters, opinions, news, personal views and commentary were excluded. The studies related to CQ and HCQ use against COVID-19, as well as studies involving their mechanisms of action against COVID-19 were included. Additionally, an in silico study (computer simulation) about the mechanism of action of CQ and HCQ against SARS-CoV-2 was included. In vitro and in vivo experimental studies describing the effect of CQ and HCQ on coronavirus were added. Clinical studies and case reports involving CQ and HCQ administration to treat SARS-CoV-2 were also included.

Review and Results

Reviews

Regarding the review articles, some of them are favorable for HCQ/CQ use, whereas other reviews are more concerned about their use. Some reviews presented the same favorable clinical and in vitro studies, due to the low number of studies already approved and published. In other words, when the review articles consider that either CQ or HCQ are effective, in fact they were based on studies with a lack of evidence for any conclusion. Additionally, there is a review that provides studies about the mechanism of action of CQ and HCQ against COVID-19.

In silico Study

The in silico study (27) simulated where CQ and HCQ can act on viral replication cycle as possible candidates for the treatment against the SARS-CoV-2, before their clinical evaluation. The authors observed that CQ and HCQ were able to prevent the binding of the viral protein to gangliosides, a potential domain involved in the attachment of the virus to human cells. However, in order to verify CQ and HCQ efficacy, these drugs need careful in vivo study before clinical evaluation.

In vitro Studies

Among the selected studies, four articles of in vitro studies were from previous uses of CQ/HCQ not related to COVID-19 (14,29–31). However, these studies presented other types of coronavirus, and were included in order to verify the use of HCQ/CQ. Regarding the COVID-19 outbreak, only one in vitro study (28) specifically addressed CQ/HCQ use. Taken together, all in vitro studies demonstrated a great potential of CQ and HCQ against the coronavirus. Yao et al. 2020 (28) demonstrated that HCQ in vitro was more potent than CQ; according to PBPK (physiologically based pharmacokinetics) modelling results, HCQ dose was 400mg twice a day, followed by a maintenance of 200mg, twice a day, for 4 days; CQ dose was 500 mg twice per 5 days.

Clinical Studies

All clinical studies included in Table 1 presented some methodology shortcomings. For example, Chen et al. 2020 (26) did not specify other drugs and doses used besides HCQ (dose = 400 mg/d) during the treatment. Thus, there was no specific treatment for control groups. Gautret et al. 2020 (23) also had issues with control groups; the authors did not provide any information about the control group treatment. Besides, each study group was treated in different places. Additionally, all studies presented a reduced number of enrolled patients (ranged from 13 to 62). Furthermore, almost all studies attributed patient survival to CQ/HCQ use and patient death to lack of medication and/or disease complications. Indeed, studies only demonstrated the improvement of medication use against COVID-19, with some patients discontinued due to severe adverse drug reactions (ADR) or death.

Pharmacokinetic Considerations

CQ and HCQ have been used as standard for malaria for more than 40 years, but the rising of resistant parasites in malaria endemic regions reduced their use (58). Besides the antimalarial employment, they have also been used in oral therapy of choice for cutaneous and systemic lupus erythematosus and rheumatoid arthritis.

All the established data for both drugs were obtained from previous studies that showed well described applications such as in malaria or lupus. The gastrointestinal absorption of CQ and HCQ is excellent, favored by the water-soluble properties of these molecules (0.7-0.8 bioavailability). This absorption is favored by the concomitant intake of food and can be altered by severe malnutrition (59). The maximum plasma concentrations are obtained within 1 to 2 hours after HCQ administration and within 2 to 6 hours for CQ. It should be noted that the bioavailability of these

molecules, particularly HCQ, can vary significantly from one subject to another (59–61).

Both CQ and HCQ are widely distributed in the body, especially in red blood cells with regard to CQ, and in the liver and kidney with regard to HCQ. CQ is extensively distributed with a large total apparent volume of distribution (Vd), higher than 100 L/kg. Because of its Vd, distribution rather than elimination processes determine the blood concentration profile of CQ in patients (62).

These two molecules have affinity for cells containing melanin, which partially explain some adverse effects (particularly ocular), but they also bind to mononuclear cells, muscles, etc. CQ and HCQ pass weakly into breast milk (63,64). In plasma, protein binding ranges between 30 and 40% with binding to both albumin and alpha 1 glycoprotein (59).

CQ is metabolized to an active metabolite (N-desethylchloroquine) and other metabolites primarily by CYP2C8 and to a lesser extent by CYP3A4 and CYP2D6 (65,66). There is an association between CYP2C8*2 and *3 and gametocytemia and parasitemia low clearance rates in CQ/primaquine treated patients, but with low relevance (67). CYP2D6 is also responsible for metabolism of CQ, and polymorphisms for this isoform have already been described in literature (68). In vitro and in vivo, CQ and desethylchloroquine competitively inhibit CYP2D1/6-mediated reactions (69).

HCQ is partially metabolized in the liver before being eliminated by the kidney (59). Elimination is mainly renal, presenting values ranging from 50 to 60%. A large amount of the administered dose of CQ is found in the urine, mainly in unchanged form. The plasma half-life of these molecules is long: 10 to 30 days for CQ and 20 to 40 days for HCQ. These half-lives vary widely depending on the patients and the daily doses received. Note, it will take several weeks to reach steady-state concentrations.

Renal CQ clearance accounts for about half of its total systemic clearance. CQ exhibits complex pharmacokinetics in adults and children, so that plasma levels of the drug shortly after its administration are determined by the rate of distribution and not by the rate of elimination. Due to extensive attachment to tissues, a loading dose is required to obtain effective plasma concentrations. This way, the interindividual variation is related to distribution including any factor that can potentially influence the absorption, metabolism or elimination of the drug. These include: age, body mass index, compliance, other medications, dose, intestinal or renal or liver disease and smoking (62).

For malaria, lupus and rheumatoid arthritis there is a linear correlation between clinical response and blood concentration. Dosing schedules for malaria treatment usually gives higher doses, for 1 up to 3 or 5 days. A pharmacokinetically suitable regimen is to administer an initial dose of 10 mg of base/kg, followed by 5 mg/kg, 6 to 8 hours later and 5 mg/kg on each of the following 2 days. Another more practical regimen, used in many areas, consists of 10 mg/kg on the first day, followed by 7.5mg/kg on the second and third days (59,69).

The data for the use during COVID-19 pandemic are few and includes little patients. Simulations about data obtained in literature make possible predictions in a mechanistic PK/virologic/QT model for HCQ to predict SARS-CoV-2 rate of viral decline and QT prolongation. SARS-CoV-2 viral decline was associated with HCQ pharmacokinetics ($P < 0.001$). The extrapolated patient EC50 was 4.7 μM, related to doses higher than 400mg, twice a day, were predicted to rapidly decrease viral loads, reducing the proportion of patients with detectable infection, and shortening treatment courses, compared to lower dose (≤400 mg daily) regimens. The big concern is that higher doses were also predicted to prolong QT intervals, pointing to important clinical implications. Due to COVID-19's variable natural history, lower dose regimens may not present efficacy, making results in treated groups indistinguishable from controls (70).

CQ and HCQ: ADRs and Safety Outcome

The therapeutic traditional uses of CQ and HCQ for the treatment of malaria, rheumatoid arthritis and systemic lupus erythematosus may be accompanied by several adverse effects of varying degrees of severity. Before detailing the most important drug-related adverse effects, it is important to describe some general terms.

ADR is a harmful and unintended response to a drug that occurs at therapeutic doses normally used in humans for prophylaxis or treatment. An ADR is the clinical manifestation of a drug treatment that occurs when a substance (a parent drug or a metabolite, a contaminant, etc.) is distributed through the body tissues and interacts with a macromolecule (a receptor or an enzyme), resulting in a physiological or pathological change, i.e. an adverse effect (71–74).

Regarding the adverse effects, a toxic effect is an adverse effect arising from supratherapeutic concentrations, i.e. an exaggeration of the desired pharmacologi-

cal effect, usually related to genetic polymorphisms, age, sex, drug interactions and some diseases, like renal and hepatic diseases. A collateral (or side) effect occurs at therapeutic concentrations and may be caused by a pharmacological mechanism other than the therapeutic action, or throughout the same pharmacological action, but in a different tissue. Hypersusceptibility reaction is a general term that describes adverse effects at subtherapeutic concentrations in susceptible patients (72,73).

Although there are several in vitro studies on either CQ or HCQ efficacy against SARS-CoV-2, the required high doses, necessary to achieve a high concentration, may cause toxicity in humans (75). In this study, we will emphasize both collateral and toxic effects of these drugs.

General Adverse Effects

CQ and HCQ have several ocular and systemic adverse effects, which include effects in the gastrointestinal tract, neurological, neuromuscular, dermatological and cardiological systems (76–79). Ponchet et al. (2005) (78) evaluated 350 patients with systemic lupus erythematosus treated with CQ diphosphate (250 mg/day). The authors observed adverse effects in 35.7% of the patients: 17.4% ocular, 10% gastrointestinal, 3.4% dermatologic, 2.9% headache, 1.7% neuromuscular and 0.3% psychiatric effects. Braga et al. (2015) (77) assessed 50 patients with P. vivax malaria before and after treatment with CQ (10 mg/kg/day during 3 days) and primaquine (30 mg/day during 7 days or 15 mg/day during 14 days). The adverse effects related were blurred vision 54%, meso or hypogastric pain 32%, nausea 32%, diarrhea 24%, lack of appetite 24%, vomiting 22%, bitter taste in the mouth 40%, pruritus 22%, "stinging" skin 22%, paresthesia 6%, choluria 44%, pale stools 12%, weakness/malaise 36%, and insomnia 46%. The studies mentioned show that gastrointestinal and dermatological reactions are the most common systemic adverse effects, while cardiac disorders are rare when CQ is used in accordance with guidelines for the rational use of medicines. Nevertheless, it is important to note that cardiac effects involve high morbidity and mortality. Higher doses contribute considerably to enhance adverse effects frequency and severity.

Cardiac arrhythmias

Wagner et al. (2010) (80) suggested that CQ-induced cardiotoxicity occurs by blocking open-channel of fast transient outward K+ current (Ito). Cardiac reactions are characterized by conduction disorders, such as QT-interval prolongation and atrioventricular block; and cardiomyopathy, such as hypertrophy, and congestive heart failure. In 2012, Tönnesmann et al. (2012) (81) questioned whether cardiac damage by CQ was rare or underreported because it may be asymptomatic for a long period. Studies have shown that conduction disorders precede cardiomyopathy and CQ treatment continues even in the presence of these reactions, due to inappropriate evaluation. There is still a lack of studies evaluating CQ-induced cardiotoxicity (82).

A systematic review evaluated eighty-six individual cases or short series, with a total of 127 patients treated with CQ, HCQ, or both in succession, during a long time (median 7 years; cumulative dose of 803g and 1235g for HCQ). The authors reported that 85% of the patients had conduction disorders, 22% hypertrophy, 9.4% hypokinesia, 26.8% heart failure, 3.9% pulmonary arterial hypertension, and 7.1% valvular dysfunction. Withdrawal of therapy was described for 78 patients, of which 44.9% recovered cardiac functions, 12.9% presented irreversible damage and 30.8% died (82).

Recently, the French National Agency for Medicines and Health Products Safety (ANSM - L'Agence Nationale de Sécurité du Médicament et des Produits de Santé), in collaboration with pharmacovigilance centers, reported that 82 from about 100 patients infected with COVID-19, showed severe ADR, using lopinavir-ritonavir and HCQ for treatment, including 4 cases of death. The effects observed were hepatotoxicity, nephrotoxicity, retinal damage, and cardiovascular reactions. In a second study with 53 cases of cardiac adverse effects (43 cases with HCQ alone or in combination, mainly with azithromycin), ANSM revealed 7 cases of sudden death (3 were recovered by external electric shock) and 12 cardiac disorders, such as electrocardiographic rhythm, and conduction disorders that include QT-interval prolongation (83).

Borba et al. (2020) (84) suspended a study with 440 patients enrolled, because 81 of them developed severe ADR, mainly cardiac reactions. The authors evaluated high (600 mg) and low doses (450 mg) of CQ in hospitalized patients with SARS. The patients also received ceftriaxone and azithromycin. It was observed that QT-interval prolongation was more pronounced in patients receiving high doses of CQ, and total fatality rate was 13.5%.

Ocular Toxicity

Both CQ and HCQ most important collateral effects are keratopathy and retinopathy. In keratopathy,

corneal deposits mainly composed of antimalarial salts are found in the basal epithelium. It may be detected in the beginning of therapy, usually after 3 days. Although most patients are asymptomatic, some of them occasionally see halos around light. Keratopathy can be reversed and usually disappears after cessation of therapy, regardless of age, sex, length of treatment and cumulative dose (85,86).

Rare retinopathy is one the most serious CQ and HCQ-associated adverse effects. Drug-induced retinopathy is an irreversible and usually progressive phenomenon, related to the total amount administered, i.e. cumulative dose, and long-term use. First changes are observed in ganglion cells cytoplasm, including retina photoreceptors degeneration. Furthermore, both CQ and HCQ have a selective affinity for melanin, located in the retinal pigment epithelium, impairing retinal metabolism. The drug may be stored within this tissue for years, even after therapy discontinuation (87–89). Although CQ damages both inner and outer retina, HCQ does not seem to harm significantly the inner retina (90).

Clinically, drug induced-retinopathy causes the atrophy of retinal pigment epithelium, leading to a bilateral damage in macular pigment, namely "bull's eye" maculopathy. This atrophy, together with neurosensory retina atrophy, may occur in the advanced stages of the retinal toxicity. Patients usually report central vision loss, visual field impairments, color vision deficiency and night blindness (91,92). Whereas up to 25% of patients reported ocular adverse effects caused by CQ use, for HCQ this number reached up to 3.5% (89).

According to the American Academy of Ophthalmology, risk factors for drug-induced retinopathy include: daily dose greater than 2.3mg of CQ/kg of bodyweight or 5.0mg of HCQ/kg of bodyweight; duration of therapy greater than 5 years; high percentage of body fat; renal or hepatic disease; over 60 years old; and genetic factors (89,90).

As detailed in the pharmacokinetic section, both CQ and HCQ are primarily stored in melanotic tissues, liver and kidneys, and it is slowly eliminated from the body. Thus, to prevent serious ocular adverse effects, recommended daily doses should be equal to or less than: 250mg for CQ and 750 mg for HCQ. Alongside dose, a long-term duration of therapy may increase the risk of retinopathy, even post-cessation. Some reports describe delayed reactions, a decade after long-term CQ administration (93).

As both drugs are not stored in lipid tissues, real bodyweight formulas for dose calculation may cause toxic effects in obese individuals. Hepatic and renal diseases may compromise both CQ and HCQ biotransformation and excretion processes, resulting in a toxic effect. Age-related general changes in the drug disposition mechanisms of elderly patients may lead to toxic effects. Moreover, as they usually have a macular degeneration, the risk of retinopathy in healthy retina tissue increases (89,90).

It is hypothesized that CYP2C8, CYP3A4 and CYP2D6 poor metabolizers may negatively impact CQ biotransformation, causing a toxic effect. However, both renal excretion and multiple enzymes involved in CQ biotransformation largely contribute to drug elimination processes, reducing CYP genetic polymorphisms' negative impact (67).

COVID-19: CQ, HCQ, Concurrent Medications and ADRs

Many concurrent medications can affect the pharmacological activity of a drug by either decreasing or increasing its action. This topic focuses on drug-drug interactions through which a drug can change another drug absorption, distribution, biotransformation and/or excretion, usually modifying its total plasma concentration, i.e. a pharmacokinetic interaction; or modify another drug mechanism of action, i.e. a pharmacodynamic interaction (94).

Several patients have received off-label drug therapies for COVID-19 treatment. Among them, we will discuss the possible interactions between CQ or HCQ combined with ritonavir-lopinavir or azithromycin (95).

The antiretroviral therapy with ritonavir-lopinavir combination profoundly inhibits CYP3A and CYP2D6 activities, impairing drug clearance. This could increase the affected drug total plasma concentration as well as the area under the curve, leading to a toxic effect (96). Although there is no scientific information regarding the interaction between CQ or HCQ combined with ritonavir-lopinavir, it may result in higher CQ or HCQ plasma concentrations, increasing cardiac arrhythmias risk.

The main concern on azithromycin use is its arrhythmogenic potential, a risk already described with another macrolide: erythromycin. Ventricular arrhythmias are related to the QT-interval prolongation, causing fast and chaotic heartbeat, which may result in a cardiac arrest and sudden death (97). The combination between azithromycin and CQ or HCQ increases the risk of ventricular arrhythmia, especially torsades de

pointes, due to an additive effect caused by the QT-interval prolongation. Thus, this co-administration should be carefully monitored or avoided (98).

Discussion

Summary of evidence

CQ and HCQ appear to block viral entry into cells, inhibiting host receptor glycosylation, proteolytic processing and endosomal acidification. HCQ in vitro appears to have a more potent antiviral activity than CQ (28). However, this same effect was previously reported for Zika virus infection and it was later shown to be ineffective for treatment and potentially harmful to patients (99). Indeed, in vitro studies have already demonstrated an antiviral effect including against the coronavirus. However, these results have never been able to introduce CQ or HCQ as an antiviral agent in any guideline of any society. The reason for this is the lack of the same results in experimental or clinical studies. This corroborates to the fact that, to date, there is insufficient data to confirm or deny that HCQ or CQ should be safely used for COVID-19 treatment.

In particular, although extremely rare, these agents can prolong the QT interval and although well tolerated in most patients, should be avoided or used with caution and with careful monitoring in patients with a prolonged baseline QT interval or associated with other agents that affect cardiac conduction. In addition, medications may be associated with other risks, such as retinopathy (extremely long term dosing) or rarely cardiomyopathy. Although serious adverse effects are highly uncommon and these drugs have been used for decades with relative safety for other pathologies, off label use for the treatment of COVID-19 has become common even though prospective double blind studies are lacking. Studies related to early dosing for COVID-19 or prophylaxis are ongoing. When these drugs are considered for COVID-19, many questions clearly remain unanswered: When to start using them? What dose should be prescribed? How long should it be prescribed? Which patients can benefit from its use? Which patients should not be prescribed? Should it be used concurrently with any other drugs? In this review, we repeat all these questions. We suggest the new studies should focus their objectives on answering these questions. The search for any medical treatment must be guided exclusively by research and conducted according to well-established scientific methods, with clear protocols and subordinated to ethical values and peer reviews. Medicines such as HCQ have been used effectively in patients with malaria and autoimmune diseases. However, we hope future studies will answer these important questions as to whether these medications can be useful in the treatment of COVID-19.

Limitations

There were some limitations to this review. The majority of articles found were letters to editors, news, hot point, point of view, which were excluded from this review. As a result of this pandemic, it is important to emphasize that new studies have been published every day regarding this subject, most of them with methodological shortcomings. The majority of in vitro studies found were not related to COVID-19; these papers were related to other types of coronavirus. The few number of clinical studies presented shortcomings in methodology.

Conclusions

In conclusion, at this review article, based on studies that have been produced from evidence to date, it is not possible to precisely state the benefits of CQ and/or HCQ in patients presenting with COVID-19. Whether there is a clinical role for these agents, in particular early in the disease process, will require additional studies in the future.

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References

1. Lai CC, Shih TP, Ko WC, Tang HJ, Hsueh PR. Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and coronavirus disease-2019 (COVID-19): The epidemic and the challenges. *Int J Antimicrob Agents* 2020; 55:105924.
2. Zhou P, Yang X-L, Wang X-G, et al. Discovery of a novel coronavirus associated with the recent pneumonia outbreak in humans and its potential bat origin. *Nature* 2020.
3. Kannan S, Shaik Syed Ali P, Sheeza A, Hemalatha K. COVID-19 (Novel Coronavirus 2019) - recent trends. *Eur Rev Med Pharmacol Sci* 2020; 24:2006-2011.
4. World Health Organization - WHO. WHO Timeline - COVID-19. 2020. https://www.who.int/news-room/detail/27-04-2020-who-timeline---covid-19. Accessed March 11, 2020.
5. Wu Z, McGoogan JM. Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention. *JAMA* February 2020. (Epub ahead of print)
6. Jin Y, Yang H, Ji W, et al. Virology, epidemiology, pathogenesis, and control of covid-19. *Viruses* 2020; 12:372.
7. Akhmerov A, Marbán E. COVID-19 and the Heart. *Circ Res* 2020; 126:1443-1455.
8. Touret F, de Lamballerie X. Of chloroquine and COVID-19. *Antiviral Res* 2020; 177:104762.
9. Schrezenmeier E, Dörner T. Mechanisms of action of hydroxychloroquine and chloroquine: implications for rheumatology. *Nat Rev Rheumatol* 2020; 16:155-166.
10. Inglot AD. Comparison of the antiviral activity in vitro of some non-steroidal anti-inflammatory drugs. *J Gen Virol* 1969; 4:203-214.
11. Rolain J-M, Colson P, Raoult D. Recycling of chloroquine and its hydroxyl analogue to face bacterial, fungal and viral infections in the 21st century. *Int J Antimicrob Agents* 2007; 30:297-308.
12. Wang M, Cao R, Zhang L, et al. Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Res* 2020; 30:269-271.
13. Kwiek JJ, Haystead TAJ, Rudolph J. Kinetic Mechanism of Quinone Oxidoreductase 2 and Its Inhibition by the Antimalarial Quinolines. *Biochemistry* 2004; 43:4538-4547.
14. Vincent MJ, Bergeron E, Benjannet S, et al. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virol J* 2005; 2:69.
15. Infectious Diseases Society of America. Infectious Diseases Society of America Guidelines on the Treatment and Management of Patients with COVID-19. https://www.idsociety.org/practice-guideline/covid-19-guideline-treatment-and-management/. Published 2020. Accessed April 16, 2020.
16. Grein J, Ohmagari N, Shin D, et al. Compassionate Use of Remdesivir for Patients with Severe Covid-19. *N Engl J Med* April 2020.
17. Ferrey AJ, Choi G, Hanna RM, et al. A Case of Novel Coronavirus Disease 19 in a Chronic Hemodialysis Patient Presenting with Gastroenteritis and Developing Severe Pulmonary Disease. *Am J Nephrol* March 2020; 51:337-342.
18. Spezzani V, Piunno A, Iselin H-U. Benign COVID-19 in an immunocompromised cancer patient - the case of a married couple. *Swiss Med Wkly* April 2020.
19. Inciardi RM, Lupi L, Zaccone G, et al. Cardiac Involvement in a Patient With Coronavirus Disease 2019 (COVID-19). *JAMA Cardiol* March 2020.
20. Bartiromo M, Borchi B, Botta A, et al. Threatening drug-drug interaction in a kidney transplant patient with Coronavirus Disease 2019 (COVID-19). *Transpl Infect Dis* April 2020.
21. Jun C, Danping L, Li L, et al. A pilot study of hydroxychloroquine in treatment of patients with common coronavirus disease-19 (COVID-19). *J Zhejiang Univ Medical Sci* 2020; 49(1).
22. Xu X-W, Wu X-X, Jiang X-G, et al. Clinical findings in a group of patients infected with the 2019 novel coronavirus (SARS-Cov-2) outside of Wuhan, China: retrospective case series. *BMJ* 2020; 368:m606.
23. Gautret P, Lagier J-C, Parola P, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int J Antimicrob Agents* March 2020.
24. Perinel S, Launay M, Botelho-Nevers É, et al. Towards Optimization of Hydroxychloroquine Dosing in Intensive Care Unit COVID-19 Patients. *Clin Infect Dis* April 2020.
25. Huang M, Tang T, Pang P, et al. Treating COVID-19 with Chloroquine. *J Mol Cell Biol* 2020; 12:322-325.
26. Chen Z, Hu J, Zhang Z, et al. Efficacy of hydroxychloroquine in patients with COVID-19: results of a randomized clinical trial. *MedRxiv* 2020.
27. Fantini J, Di Scala C, Chahinian H, Yahi N. Structural and molecular modelling studies reveal a new mechanism of action of chloroquine and hydroxychloroquine against SARS-CoV-2 infection. *Int J Antimicrob Agents* April 2020.
28. Yao X, Ye F, Zhang M, et al. In Vitro Antiviral Activity and Projection of Optimized Dosing Design of Hydroxychloroquine for the Treatment of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). *Clin Infect Dis* March 2020.
29. Keyaerts E, Vijgen L, Maes P, Neyts J, Van Ranst M. In vitro inhibition of severe acute respiratory syndrome coronavirus by chloroquine. *Biochem Biophys Res Commun* 2004; 323:264-268.
30. Keyaerts E, Li S, Vijgen L, et al.

Antiviral Activity of Chloroquine against Human Coronavirus OC43 Infection in Newborn Mice. *Antimicrob Agents Chemother* 2009; 53:3416-3421.

31. Barnard DL, Day CW, Bailey K, et al. Evaluation of immunomodulators, interferons and known in vitro SARS-coV inhibitors for inhibition of SARS-coV replication in BALB/c mice. *Antivir Chem Chemother* 2006; 17:275-284.
32. Devaux CA, Rolain J-M, Colson P, Raoult D. New insights on the antiviral effects of chloroquine against coronavirus: what to expect for COVID-19? *Int J Antimicrob Agents* March 2020.
33. Du Y-X, Chen X-P. Favipiravir: pharmacokinetics and concerns about clinical trials for 2019-nCoV infection. *Clin Pharmacol Ther* April 2020.
34. Gbinigie K, Frie K. Should chloroquine and hydroxychloroquine be used to treat COVID-19? A rapid review. *BJGP Open* April 2020.
35. Kakodkar P, Kaka N, Baig MN. A comprehensive literature review on the clinical presentation, and management of the Pandemic Coronavirus Disease 2019 (COVID-19). *Cureus* 2020; 12:e7560.
36. Shah S, Das S, Jain A, Misra DP, Negi VS. A systematic review of the prophylactic role of chloroquine and hydroxychloroquine in Coronavirus Disease-19 (COVID-19). *Int J Rheum Dis* April 2020.
37. Cortegiani A, Ingoglia G, Ippolito M, Giarratano A, Einav S. A systematic review on the efficacy and safety of chloroquine for the treatment of COVID-19. *J Crit Care* March 2020.
38. Lai C-C, Liu YH, Wang C-Y, et al. Asymptomatic carrier state, acute respiratory disease, and pneumonia due to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2): Facts and myths. *J Microbiol Immunol Infect* March 2020.
39. Kapoor A, Pandurangi U, Arora V, et al. Cardiovascular risks of hydroxychloroquine in treatment and prophylaxis of COVID-19 patients: A scientific statement from the Indian Heart Rhythm Society. *Indian Pacing Electrophysiol J* April 2020.
40. Pereira BB. Challenges and cares to promote rational use of chloroquine and hydroxychloroquine in the management of coronavirus disease 2019 (COVID-19) pandemic: a timely review. *J Toxicol Environ Health B Crit Rev* 2020; 23:177-181.
41. Haşlak F, Yıldız M, Adrovic A, Barut K, Kasapçopur Ö. Childhood rheumatic diseases and COVID-19 pandemic: An intriguing linkage and a new horizon. *Balkan Med J* 2020; 37:184-188.
42. Singh AK, Singh A, Shaikh A, Singh R, Misra A. Chloroquine and hydroxychloroquine in the treatment of COVID-19 with or without diabetes: A systematic search and a narrative review with a special reference to India and other developing countries. *Diabetes Metab Syndr Clin Res Rev* 2020; 14:241-246.
43. Gupta N, Agrawal S, Ish P. Chloroquine in COVID-19: the evidence. *Monaldi Arch Chest Dis* 2020;90(1).
44. Rosa SGV, Santos WC. Clinical trials on drug repositioning for COVID-19 treatment. *Pan Am J public Heal* 2020; 44:e40.
45. Gupta R, Misra A. Contentious issues and evolving concepts in the clinical presentation and management of patients with COVID-19 infectionwith reference to use of therapeutic and other drugs used in Co-morbid diseases (Hypertension, diabetes etc). *Diabetes Metab Syndr* 2020; 14:251-254.
46. Zhang C, Huang S, Zheng F, Dai Y. Controversial treatments: An updated understanding of the coronavirus disease 2019. *J Med Virol* March 2020.
47. Dashraath P, Jing Lin Jeslyn W, Mei Xian Karen L, et al. Coronavirus Disease 2019 (COVID-19) Pandemic and Pregnancy. *Am J Obstet Gynecol* March 2020.
48. Hussain A, Bhowmik B, do Vale Moreira NC. COVID-19 and diabetes: Knowledge in progress. *Diabetes Res Clin Pract* 2020; 162.
49. Zhou D, Dai S-M, Tong Q. COVID-19: a recommendation to examine the effect of hydroxychloroquine in preventing infection and progression. *J Antimicrob Chemother* March 2020.
50. Liu W, Li H. COVID-19: Attacks the 1-Beta Chain of Hemoglobin and Captures the Porphyrin to Inhibit Human Heme Metabolism. *Chemrxiv* 2020.
51. Plantone D, Koudriavtseva T. Current and Future Use of Chloroquine and Hydroxychloroquine in Infectious, Immune, Neoplastic, and Neurological Diseases: A Mini-Review. *Clin Drug Investig* 2018; 38:653-671.
52. Agrawal S, Goel AD, Gupta N. Emerging prophylaxis strategies against COVID-19. *Monaldi Arch Chest Dis* 2020; 90(1).
53. Xie M, Chen Q. Insight into 2019 novel coronavirus — an updated intrim review and lessons from SARS-CoV and MERS-CoV. *Int J Infect Dis* April 2020.
54. Sanders JM, Monogue ML, Jodlowski TZ, Cutrell JB. Pharmacologic treatments for coronavirus disease 2019 (COVID-19): A review. *JAMA* April 2020.
55. Lu C-C, Chen M-Y, Chang Y-L. Potential therapeutic agents against COVID-19. *J Chinese Med Assoc* April 2020:1.
56. Juurlink DN. Safety considerations with chloroquine, hydroxychloroquine and azithromycin in the management of SARS-CoV-2 infection. *Can Med Assoc J* April 2020.
57. Zhai P, Ding Y, Wu X, Long J, Zhong Y, Li Y. The epidemiology, diagnosis and treatment of COVID-19. *Int J Antimicrob Agents* 2020; 55:105955.
58. Mengue JB, Held J, Kreidenweiss A. AQ-13 - an investigational antimalarial drug. *Expert Opin Investig Drugs* 2019; 28:217-222.
59. Furst DE. Pharmacokinetics of hydroxychloroquine and

chloroquine during treatment of rheumatic diseases. *Lupus* 1996; 5:S11-S15.

60. McLachlan AJ, Tett SE, Cutler DJ, Day RO. Absorption and in vivo dissolution of hydroxycholoroquine in fed subjects assessed using deconvolution techniques. *Br J Clin Pharmacol* 1993; 36:405-411.
61. Tett S, Day R, Cutler D. Hydroxychloroquine relative bioavailability: Within subject reproducibility. *Br J Clin Pharmacol* 1996; 41:244-246.
62. Krishna S, White NJ. Pharmacokinetics of Quinine, Chloroquine and Amodiaquine. *Clin Pharmacokinet* 1996; 30:263-299.
63. Akintonwa A, Gbajumo SA, Biola Mabadeje AF. Placental and Milk Transfer of Chloroquine in Humans. *Ther Drug Monit* 1988; 10:147-149.
64. Nation RL, Hackett LP, Dusci LJ, Ilett KF. Excretion of hydroxychloroquine in human milk [letter]. *Br J Clin Pharmacol* 1984; 17:368-369.
65. Projean D, Baune B, Farinotti R, et al. In vivo metabolism of chloroquine:identification of CYP2C8, CYP3A4 and CYP2D6 as main isoforms catalyzing n-desethychloroquine formation. *Drug Metab Dispos* 2003; 31:748-754.
66. Sortica VA, Lindenau JD, Cunha MG, et al. The effect of SNPs in CYP450 in chloroquine/primaquine Plasmodium vivax malaria treatment. *Pharmacogenomics* 2016; 17:1903-1911.
67. Gil J, Gil Berglund E. CYP2C8 and antimalaria drug efficacy. *Pharmacogenomics* 2007; 8(2):187-198.
68. Elewa H, Wilby KJ. A Review of Pharmacogenetics of Antimalarials and Associated Clinical Implications. *Eur J Drug Metab Pharmacokinet* 2017; 42:745-756.
69. Ducharme J, Farinotti R. Clinical Pharmacokinetics and Metabolism of Chloroquine. *Clin Pharmacokinet* 1996; 31:257-274.
70. Garcia-Cremades M, Solans BP, Hughes E, et al. Optimizing hydroxychloroquine dosing for patients with COVID-19: An integrative modeling approach for effective drug repurposing. *Clin Pharmacol Ther* 2020; 108:253-263
71. WHO. International drug monitoring: the role of national centres. Report of a WHO meeting. *World Health Organ Tech Rep Ser* 1972;498:1-25.
72. Aronson JK, Ferner RE. Joining the DoTS: New approach to classifying adverse drug reactions. *Br Med J* 2003; 327:1222-1225.
73. Aronson JK, Ferner RE. Clarification of terminology in drug safety. *Drug Saf* 2005; 28:851-870.
74. Aronson JK. Distinguishing hazards and harms, adverse drug effects and adverse drug reactions: Implications for drug development, clinical trials, pharmacovigilance, biomarkers, and monitoring. *Drug Saf* 2013; 36:147-153.
75. Kupferschmidt K, Cohen J. Race to find COVID-19 treatments accelerates. *Science (80-)* 2020; 367:1412-1413.
76. Ponticelli C, Moroni G. Hydroxychloroquine in systemic lupus erythematosus (SLE). *Expert Opin Drug Saf* 2017; 16:411-419.
77. Braga CBE, Martins AC, Cayotopa ADE, et al. Side effects of chloroquine and primaquine and symptom reduction in malaria endemic area (Mâncio lima, Acre, Brazil). *Interdiscip Perspect Infect Dis* 2015; 2015:1-7.
78. Ponchet MRNC, Vilela MAC, Suni Sinahara KK, De Freitas Dotto P. Evaluation of adverse events triggered by the use of chloroquine diphosphate, emphasis on retina toxicity, in 350 patients with lupus erythematosus. In: *Anais Brasileiros de Dermatologia* 2005; 80:275-282.
79. Klinger G, Morad Y, Westall CA, et al. Ocular toxicity and antenatal exposure to chloroquine or hydroxychloroquine for rheumatic diseases. *Lancet* 2001; 358:813-814.
80. Wagner M, Riepe KG, Eberhardt E, Volk T. Open channel block of the fast transient outward K+ current by primaquine and chloroquine in rat left ventricular cardiomyocytes. *Eur J Pharmacol* 2010; 647:13-20.
81. Tönnesmann E, Stroehmann I, Kandolf R, et al. Cardiomyopathy caused by longterm treatment with chloroquine: A rare disease, or a rare diagnosis? *J Rheumatol* 2012; 39:1099-1103.
82. Chatre C, Roubille F, Vernhet H, Jorgensen C, Pers YM. Cardiac Complications Attributed to Chloroquine and Hydroxychloroquine: A Systematic Review of the Literature. *Drug Saf* 2018; 41:919-931.
83. ANSM. Médicaments utilisés chez les patients atteints du COVID-19 : une surveillance renforcée des effets indésirables - Point d'information. https://www.ansm.sante.fr/S-informer/Actualite/Medicaments-utilises-chez-les-patients-atteints-du-COVID-19-une-surveillance-renforcee-des-effets-indesirables-Point-d-information. Published 2020. Accessed April 16, 2020.
84. Borba MGS, Val FFA, Sampaio VS, et al. Chloroquine diphosphate in two different dosages as adjunctive therapy of hospitalized patients with severe respiratory syndrome in the context of coronavirus (SARS-CoV-2) infection: Preliminary safety results of a randomized , double-blinded , phase I. *medRxiv* 2020;Preprint.
85. Easterbrook M. Ocular effects and safety of antimalarial agents. *Am J Med* 1988; 85(4 SUPPL. 1):23-29.
86. Puavilai S, Kunavisarut S, Vatanasuk M, et al. Ocular toxicity of chloroquine among Thai patients. *Int J Dermatol* 1999; 38:934-937.
87. Ventura DF, Silveira LCL, Nishi M, et al. Color vision loss in patients treated with chloroquine. *Arq Bras Oftalmol* 2003; 66(5 SUPPL.):9-15.
88. Inada ET, Watanabe KP, Tanaka SG, Sakakisbara LA. Toxicidade ocular causada pela cloroquina: Relato de caso. *Arq Bras Oftalmol* 2005; 68:407-409.
89. Lacava AC. Complicações oculares da terapêutica com a cloroquina e derivados. *Arq Bras Oftalmol* 2010; 73:384-389.
90. Marmor MF, Kellner U, Lai TYY, Melles RB, Mieler WF, Lum F. Recommendations on Screening for Chloroquine and Hydroxychloroquine Retinopathy

(2016 Revision). *Ophthalmology* 2016; 123:1386-1394.

91. Pasadhika S, Fishman GA. Effects of chronic exposure to hydroxychloroquine or chloroquine on inner retinal structures. *Eye* 2010; 24:340-346.
92. Bozinovic MST, Babic GS, Petrovic M, Karadzic J, Vulovic TA, Trenkic M. Role of optical coherence tomography in the early detection of macular thinning in rheumatoid arthritis patients with chloroquine retinopathy. *J Res Med Sci*. 2019; 24:55.
93. Kazi MS, Saurabh K, Rishi P, Rishi E. Delayed onset chloroquine retinopathy presenting 10 years after long-term usage of chloroquine. *Middle East Afr J Ophthalmol* 2013; 20:89-91.
94. Delafuente JC. Understanding and preventing drug interactions in elderly patients. In: *Critical Reviews in Oncology/Hematology*. 2003; 48:133-143.
95. Kalil AC. Treating COVID-19-off-label drug use, compassionate use, and randomized clinical trials during pandemics. *JAMA* 2020 March 24 [Online ahead of print].
96. Wyen C, Fuhr U, Frank D, et al. Effect of an antiretroviral regimen containing ritonavir boosted lopinavir on intestinal and hepatic CYP3A, CYP2D6 and P-glycoprotein in HIV-infected patients. *Clin Pharmacol Ther* 2008; 84:75-82.
97. Trifirò G, De Ridder M, Sultana J, et al. Use of azithromycin and risk of ventricular arrhythmia. *CMAJ* 2017; 189:E560-E568.
98. Karalliedde L, Clarke S, Collignon U, Karalliedde J. Adverse drug interactions: A handbook for prescribers. *Br J Clin Pharmacol* 2010; 71:301.
99. Roques P, Thiberville S-D, Dupuis-Maguiraga L, et al. Paradoxical effect of chloroquine treatment in enhancing chikungunya virus infection. *Viruses* 2018; 10:268.