Received: 19 August 2022 | Revised: 18 November 2022 | Accepted: 22 November 2022

DOI: 10.1111/cts.13468

ARTICLE

Hydroxychloroquine for treatment of non-hospitalized adults with COVID-19: A meta-analysis of individual participant data of randomized trials

Oriol Mitjà[1,2,3] | **Gilmar Reis**[4,5] | **David R. Boulware**[6] | **Adam M. Spivak**[7] | **Ammar Sarwar**[8] | **Christine Johnston**[9] | **Brandon Webb**[10] | **Michael D. Hill**[11] | **Davey Smith**[12] | **Peter Kremsner**[13,14] | **Marla Curran**[15] | **David Carter**[16] | **Jim Alexander**[15] | **Marc Corbacho**[1] | **Todd C. Lee**[6] | **Katherine Huppler Hullsiek**[6] | **Emily G. McDonald**[17] | **Rachel Hess**[7] | **Michael Hughes**[8] | **Jared M. Baeten**[9] | **Ilan Schwartz**[18] | **Luanne Metz**[11] | **Lawrence Richer**[18] | **Kara W. Chew**[19] | **Eric Daar**[20] | **David Wohl**[21] | **Michael Dunne**[15]

[1]Fight AIDS and Infectious Diseases Foundation, Barcelona, Spain

[2]Hospital Universitari Germans Trias i Pujol, Badalona, Spain

[3]Lihir Medical Center–International SOS, Lihir Island, Papua New Guinea

[4]Research Division, Cardresearch Cardiologia Assistencial e de Pesquisa, Pontifícia Universidade Católica de Minas Gerais, Bello Horizonte, Brazil

[5]Cytel Inc., Vancouver, British Columbia, Canada

[6]Division of Infectious Diseases and International Medicine, Department of Medicine, University of Minnesota, Minneapolis, Minnesota, USA

[7]University of Utah, Salt Lake City, Utah, USA

[8]Harvard Medical School, Boston, Massachusetts, USA

[9]Department of Medicine and Laboratory Medicine and Pathology, University of Washington, Seattle, Washington, USA

[10]Intermountain Health Care, University of Utah, Salt Lake City, Utah, USA

[11]University of Calgary, Calgary, Alberta, Canada

[12]Division of Infectious Diseases & Global Public Health, UC San Diego School of Medicine, San Diego, California, USA

[13]University Hospital of Tübingen, Tübingen, Germany

[14]Centre de Recherches Médicales de Lambaréné, Lambaréné, Gabon

[15]Bill & Melinda Gates Medical Research Institute, Cambridge, Massachusetts, USA

[16]MMS Holdings, Canton, Michigan, USA

[17]Division of General Internal Medicine, McGill University Health Center, Montreal, Quebec, Canada

[18]University of Alberta, Edmonton, Canada

[19]Division of Infectious Diseases, Department of Medicine, David Geffen School of Medicine at University of California, Los Angeles, California, USA

[20]Lundquist Institute at Harbor-UCLA Medical Center, Torrance, California, USA

[21]School of Medicine, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, USA

Correspondence
Oriol Mitjà, Fight AIDS and Infectious Diseases Foundation, Barcelona, Spain, Ctra. de Canyet s/n • Hosp. Univ. Germans Trias i Pujol, 08916 Badalona, Barcelona, Spain.
Email: omitja@lluita.org

Abstract

Hydroxychloroquine (HCQ) was initially promoted as an oral therapy for early treatment of coronavirus disease 2019 (COVID-19). Conventional meta-analyses cannot fully address the heterogeneity of different designs and outcomes of randomized controlled trials (RCTs) assessing the efficacy of HCQ in outpatients with mild COVID-19. We conducted a pooled analysis of individual participant data from RCTs that evaluated the effect of HCQ on hospitalization and viral load reduction in outpatients with confirmed COVID-19. We evaluated the overall treatment group effect by log-likelihood ratio test (−2LL) from a generalized linear mixed model to accommodate correlated longitudinal binary data. The analysis included data from 11 RCTs. The outcome of virological effect, assessed in 1560 participants ($N = 795$ HCQ, $N = 765$ control), did not differ significantly between the two treatment groups (−2LL = 7.66; $p = 0.18$) when adjusting for cohort, duration of symptoms, and comorbidities. The decline in polymerase chain reaction positive tests from day 1 to 7 was 42.0 and 41.6 percentage points in the HCQ and control groups, respectively. Among the 2037 participants evaluable for hospitalization ($N = 1058$ HCQ, $N = 979$ control), we found no significant differences in hospitalization rate between participants receiving HCQ and controls (odds ratio 0.995; 95% confidence interval 0.614–1.610; −2LL = 0.0; $p = 0.98$) when adjusting for cohort, duration of symptoms, and comorbidities. This individual participant data meta-analysis of 11 HCQ trials that evaluated severe acute respiratory syndrome-coronavirus 2 viral clearance and COVID-19 hospitalization did not show a clinical benefit of HCQ. Our meta-analysis provides evidence to support the interruption in the use of HCQ in mild COVID-19 outpatients to reduce progression to severe disease.

Study Highlights

WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

Various randomized-controlled trials have shown little or no efficacy of hydroxychloroquine (HCQ) for treating mild coronavirus disease 2019 (COVID-19) in the outpatient setting; however, the investigated outcomes and results are heterogeneous and cannot be fully addressed by conventional meta-analyses of aggregated data.

WHAT QUESTION DID THIS STUDY ADDRESS?

Does treatment with HCQ reduce the risk of progressing toward severe illness in individuals with mild COVID-19?

WHAT DOES THIS STUDY ADD TO OUR KNOWLEDGE?

In this meta-analysis of individual participant data, we found that HCQ does not improve viral clearance nor reduces the risk of hospitalizations when administered to individuals with mild COVID-19.

HOW MIGHT THIS CHANGE CLINICAL PHARMACOLOGY OR TRANSLATIONAL SCIENCE?

Our meta-analysis provides evidence to support the interruption in the use of HCQ in mild COVID-19 outpatients to reduce progression to severe disease.

INTRODUCTION

From the earliest days of the coronavirus disease 2019 (COVID-19) pandemic, treatment of early infection to reduce morbidity and prevent hospitalization was identified as an important area of investigation. Widespread interest quickly arose in the potential use of chemotherapeutic agents already approved for other diseases for which preliminary evidence suggested possible activity against the severe acute respiratory syndrome-coronavirus

2(SARS-CoV-2) virus.[1] Because these agents with existing indications and established safety profiles did not require investigational new drug approval, clinical trials were able to launch rapidly, providing critical platforms to investigate the potential efficacy of such "repurposed drugs" using a rigorous trial methodology.

Chloroquine and hydroxychloroquine (HCQ) are antimalarial agents that were among the first of these so-called repurposed drug candidates to attract interest in the treatment of COVID-19. These agents alkalinize the endosome and have immunomodulatory effects.[2] Preclinical data from SARS-CoV suggested in vitro inhibition of replication in cell culture, perhaps due to glycosylation of the angiotensin-converting enzyme 2 receptor or interference with endosomal transport.[3–7] Results from small, poorly designed studies lacking appropriate controls, suggested a possible clinical effect of reducing SARS-CoV-2 infection, generating substantial interest in the potential use of these agents in both scientific and lay communities.

With greater drug availability and in the context of a growing international public health emergency, HCQ was soon widely adopted in ambulatory practice and hospital treatment protocols without supporting evidence. To address this knowledge gap, clinical trials evaluating the efficacy and safety of HCQ in hospitalized and non-hospitalized COVID-19 populations were rapidly initiated. Some of these trials raised concerns regarding treatment safety in patients with COVID-19, particularly cardiovascular adverse events (e.g., conduction disorders), but also dermatological (e.g., hyperpigmentation of the skin), neuromuscular (e.g., weakness), and ophthalmological (e.g., retinopathy).[8] However, cumulative experience with HCQ showed no increased risk of adverse events in patients with COVID-19 compared with the use of this drug in approved indications, thus supporting the safe use of this drug.[9,10] Regarding efficacy, trials focusing on hospital-admitted patients were completed first, consistently demonstrating no clinical effect on the outcome of death.[11–13] Trials assessing efficacy in outpatients[14–18] or the prophylactic effect among exposed contacts also showed no benefit associated with HCQ.[19,20]

Conclusive findings in the setting of mild-COVID have been stymied, first, by the small sample size of participants in each of the multiple randomized controlled trials (RCTs), which limited statistical power, and second, by the incapacity of conventional meta-analyses to fully address heterogeneity of different trials' design and outcome measurements. In the summer of 2020, the US National Institutes of Health assembled a group of investigators conducting clinical trials studying the effect of HCQ on early, non-hospitalized patients with mild–moderate COVID-19. To strengthen the statistical power of the analyses, and allow more robust inference around well-defined clinical end points such as viral clearance and hospitalization, an individual patient data (IPD) meta-analysis was conducted using pooled data from 11 RCTs.

METHODS

Study screening and selection

The search for studies for the meta-analysis began by setting guidelines for the types of studies to be included. For eligibility assessment, studies had to be completed, enroll confirmed COVID-19 outpatients (either symptomatic or asymptomatic), include a treatment arm with 5-to-10 days HCQ at a daily dose ranging from 1600 to 4400 mg, and have a comparator arm. Individual data from participants who received HCQ combined with other drugs were excluded from the analysis.

Methods for identifying studies consisted of systematic searches on the ClinicalTrials.gov database for studies registered with the condition "COVID-19" and drug name "hydroxychloroquine" from February to October 2020. Due to the short time period since the beginning of the COVID-19 pandemic, databases indexing published journal articles were not considered.

Data extraction and quality assessment

We contacted the investigators and/or sponsors of the selected trials by email and offered them to participate in the meta-analysis of individual participant data. The datasets were shared as either coma-separated values text files or statistical analysis system datasets and mapped to study data tabulation mode-like datasets before including them into the pooled dataset. Using the integrated datasets for all studies, a participant-level dataset with baseline characteristics and population information (ADSL) was created. A dataset using AD model builder software was created for the polymerase chain reaction (PCR) test result and viral load to perform the planned analyses. Participant-level data sought included baseline information (age, sex, race, country/region, weight, height, body mass index [BMI], comorbidities, and COVID-19 characteristics) and follow-up information (PCR test results for different timepoints and hospitalization). We chose and retrieved baseline information that was relevant for subgroups analyses, such as comorbidities, symptoms, high- and low-risk groups, sampling method, collection method, assay type, and viral load at baseline. For each contributor, the data were cross-checked against the mapping spreadsheet to confirm that enough data were available as per the

requirements for analysis and end points for each respective study. All data issues, anomalies, and missing data queries were sent to contributors for their confirmation. Tables, listings, and figures were created based on the inclusion assumptions for a specific analysis.

Study end points and variables

Two co-primary end points were established: viral clearance and hospitalization rate. Viral clearance was defined as the proportion of participants with a negative PCR result for SARS-CoV-2 at the successive follow-up visits (days 3, 7, 10, and/or 14 after treatment start). To ensure consistent evaluations across all studies, the baseline was defined as the day of the first dose of the study drug. The hospitalization rate was estimated considering all hospital admissions of a patient testing positive for SARS-CoV-2 during the follow-up period (up to 30 days after treatment start). An additional analysis of viral load after HCQ administration was conducted using individual participant data from studies that had monitored viral load over time.

Other variables analyzed included demographic (age, sex, race, and country/region), clinical (weight, height, BMI, and comorbidities), and COVID-19 characteristics. Based on these characteristics, we categorized participants into low- and high-risk for severe COVID-19. High-risk criteria included age greater than or equal to 65 years, BMI greater than or equal to 30 kg/m^2, and having one of the following prespecified comorbidities: pulmonary disease, cardiovascular disease, kidney disease, immunocompromised status, liver disease, diabetes, and hypertension. Finally, we quantified treatment exposure by considering the entire treatment period (i.e., [day of the last dose]-[day of the first dose] + 1). The treatment exposure estimates for studies that did not provide a date for treatment start is described in the supplementary methods (Supplementary Material S1).

Efficacy analysis populations

A participant was considered to have a mild SARS-CoV-2 infection, and therefore met the inclusion criteria for the analysis, if they did not require hospitalization, and had a positive SARS-CoV-2 PCR test either at screening or at baseline (including those asymptomatic with positive PCR test results). The relevant days of assessment were taken directly as provided in the specific study datasets available for each specific study.

The hospitalization analysis population consisted of all participants who received at least one dose of any of the study drugs (i.e., HCQ, HCQ + azithromycin, standard of care, or placebo).

The PCR analysis population, used for the viral clearance end point, consisted of all participants who satisfied eligibility criteria, and had at least one post-baseline PCR test collected at one of the prespecified timepoints of interest (days 3, 7, 10, and 14). The viral load analysis population consisted of all participants in the PCR analysis population, who had a positive viral load quantitative measurement at both baseline and at least one post-baseline timepoint.

Statistics

Viral clearance and hospitalization data were analyzed using a generalized linear mixed model (GLMM) to accommodate for correlated longitudinal binary data. The estimated treatment effect from GLMM is the odds of a positive SARS-CoV-2 test among participants randomized to HCQ relative to control and the odds of hospitalization among participants randomized to HCQ relative to control, assessed using a log-likelihood ratio test (−2LL). The adaptive Gauss-Hermite quadrature method was used. Two statistical models (full and reduced) were fitted to assess primary objectives of whether there was evidence of any global HCQ effect. The full model accounted for both study-specific and participant-specific random effects, and fixed effects for study drug administration, study day, study drug administration-by study day-interaction, the risk variable, and duration of symptoms variable. The reduced model was similar to the full model but the fixed effects associated with study drug administration were removed. Results were presented as the odds ratio (OR) and the corresponding 95% confidence interval (CI).

The likelihood ratio test between the full and reduced models was used to assess the global null hypothesis of any HCQ treatment effect. The test statistic was compared against χ^2 distribution with degrees of freedom equal to the number of parameters in the full model minus the number of parameters in the reduced model. A one-sided test was used with nominal significance level 5%. Table S1 (Supplementary Material S1) and the statistical analysis plan (Supplementary Material S2) provide further details regarding the statistical models used for the assessment of the two primary end points.

The nominal and model-based estimated proportion of positive tests over time for each study was explored overall and by each subgroup. Analyses included the proportion of participants with SARS-CoV-2 positive and negative test results at each visit, the proportion of participants with hospitalization, and estimates of ORs for hospitalization with appropriate 95% CIs.

No formal statistical analysis was performed on the viral load data due to the limited availability of data across studies. Descriptive summary statistics were provided, showing the mean, median, and standard deviation (SD).

Subgroups for selected analyses in the aggregated populations included sex, age (<65 years, ≥65 years), risk level (low or high), and the number of days since the first symptom/s appeared (categorized as: no baseline symptoms, ≤3 days of symptoms at baseline, >3 days of symptoms at baseline). In addition to the subgroup analyses performed on all participants, a sensitivity analysis was performed post hoc to assess treatment differences when participants with negative PCR result at baseline/study day 1 were removed from the analyses.

Missing values were not imputed and were handled appropriately in the mixed model analysis. All of the model analyses were implemented using SAS PROC GLIMMIX.

RESULTS

Study characteristics

After assessing eligibility criteria, we sought data from 32 studies; 11 of them provided us with individual participant data (Figure 1). Three hundred eighty-eight participants were excluded from the meta-analysis because they had received chloroquine (trials NCT04346667 and NCT04351191), or lopinavir/ritonavir (trial NCT04403100), or HCQ plus cobicistat-boosted darunavir (trial NCT04304053). The study conducted by the Fight AIDS Foundation and Infectious Diseases (NCT04304053) had two separate cohorts: non-hospitalized adult participants with recently confirmed SARS-CoV-2 infection (FLS-1), and asymptomatic contacts exposed to a PCR-positive COVID-19 case (FLS-2).

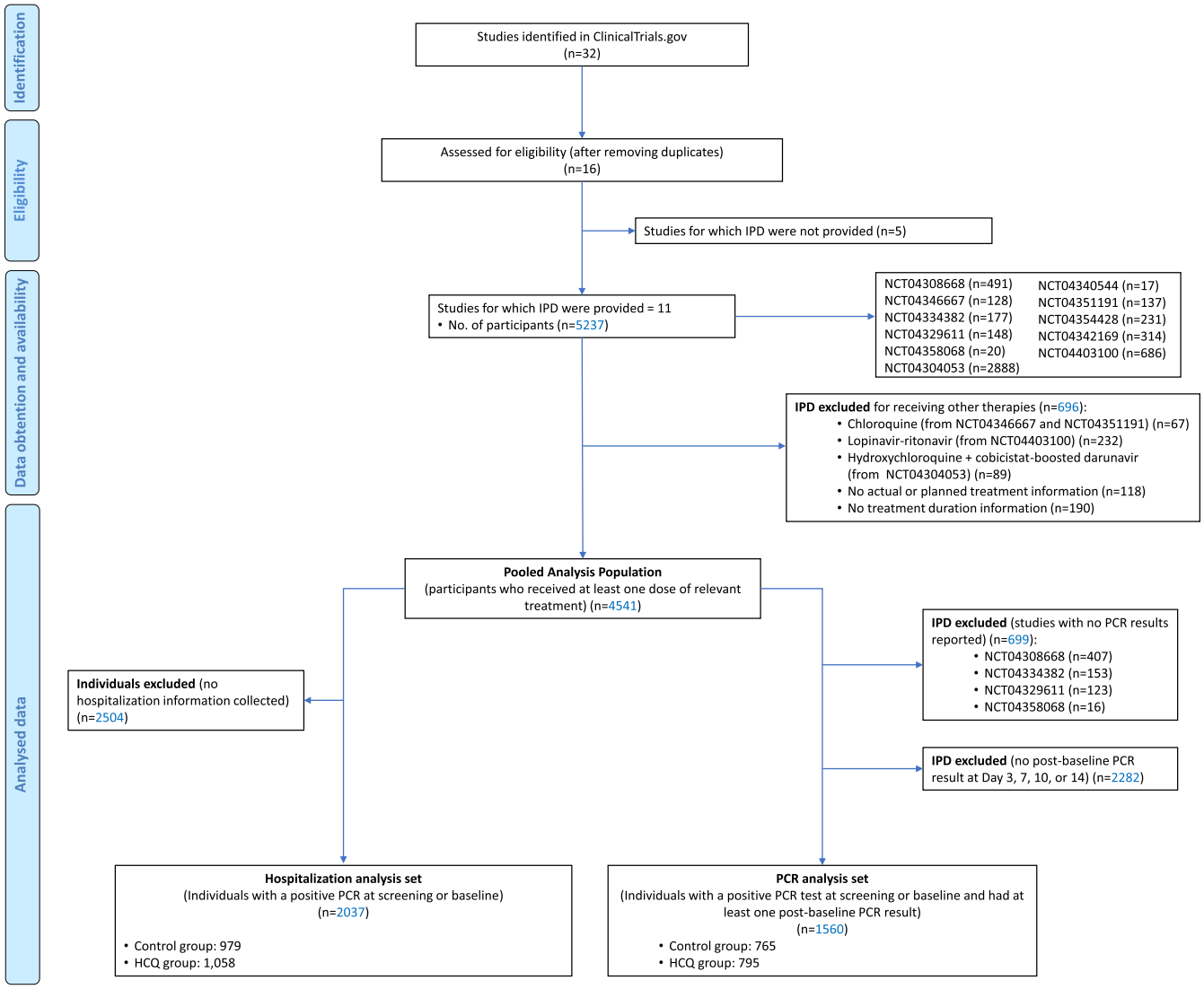

FIGURE 1 PRISMA flow-chart of the individual participant data meta-analysis. HCQ, hydroxychloroquine; IPD, individual participant data; PCR, polymerase chain reaction

TABLE 1 Overview of randomized clinical trials: design, population, investigational product, and sample size

Study CT.gov number (Institution)	Double blind	Randomized	Multicenter	Hospital outcome	Viral outcome	HCQ N[a]	Control N[a]	Population	Individual study related associated outcomes	Hydroxychloroquine dosing
1. NCT04304053 (Fight AIDS and infectious diseases foundation FLS-1 & 2, Catalonia, Spain)[18,20]		●	●	●	●	169 1225	184 1300	FLS-1: non-hospitalized adult patients with recently confirmed SARS-CoV-2 infection and less than 5 days of symptoms FLS-2: Asymptomatic contacts exposed to a PCR-positive COVID-19 case	Reduction of viral RNA load in nasopharyngeal swabs up to 7 days after the first day of study drug administration	HCQ (800 mg D1, 400 mg D2-D7) Standard of Care
2. NCT04346667 (Government of Punjab)	●	●	●	●	●	64 (32 CQ)	32	Asymptomatic after an RT-PCR proven SARS-CoV-2 infection	Two consecutive within 24 h nasopharyngeal RT-PCR at day 6 and day 7 (Primary end point)	HCQ 400 mg b.i.d. D1, D2, 200 mg b.i.d. D2-D5 HCQ 400 mg b.i.d. D1, D2, placebo b.i.d. D2-D5 CQ 500 mg b.i.d. D1-D5, placebo b.i.d. D6 Placebo CQ study drug arm not included
3. NCT04351191 (Government of Punjab)	●	●	●	●	●	69 (35 CQ)	33	Mild symptoms and RT-PCR proven SARS-CoV-2 infection	Two consecutive within 24 hours nasopharyngeal RT-PCR at day 6 and day 7 (Primary endpoint)	HCQ 400 mg b.i.d. D1, D2, 200 mg b.i.d. D2-D5 HCQ 400 mg b.i.d. D1, D2, placebo b.i.d. D2-D5 CQ 500 mg b.i.d. D1-D5, placebo b.i.d. D6 Placebo CQ study drug arm not included
4. NCT04342169 (University of Utah, USA)		●	●	●	●	158	156	Ambulatory patients with positive SARS-CoV-2 test	Duration of viral shedding, as defined by time from randomization to the first of two consecutive negative swabs, measured on days 1–14	HCQ (400 mg b.i.d. D1, 200 mg b.i.d. D2-D5) Placebo
5. NCT04354428 (University of Washington, USA)[16]	●	●	●	●	●	148	83	Men and women 18 to 80 years of age who test positive for SARS-CoV-2 within the past 72 hour in high-risk adults not requiring hospital admission	Time to clearance of nasal SARS-CoV-2, defined as 2 consecutive negative swabs (swabs collected daily Day 1 through Day 14)	HCQ 400 mg b.i.d. D1, 200 mg b.i.d. D2-D10 HCQ+ Azithromycin 400 mg b.i.d. D1, 200 mg b.i.d. D2-D10 + Azithromycin 500 mg D1, 250 mg D2-D5 Control: Ascorbic acid 500 mg b.i.d. D1, then 250 mg b.i.d. D2-D10 + folic acid 800 μg D1, 400 μg D2-D5

TABLE 1 (Continued)

Study CT.gov number (Institution)	Double blind	Randomized	Multicenter	Hospital outcome	Viral outcome	HCQ N[a]	Control N[a]	Population	Individual study related associated outcomes	Hydroxychloroquine dosing
6. NCT04358068 (NIAID via AIDS Clinical Trial Group network, USA) Results posted on ClinicalTrials.gov	●	●	●	●	●	9	11	Symptomatic, out-patient, adults (≥18 years) with SARS-CoV-2 infection	Nasopharyngeal swab at day 6 and day 20	HCQ + Azithromycin: HCQ 400 mg b.i.d. D1, 200 mg b.i.d. D2-D7 Azithromycin 500 mg D1, 250 mg D2-D5 Placebo
7. NCT04340544 (University Hospital Tübingen, Germany)	●	●	●	●	●	8	9	Adults older or equal to 18 years with positive SARS-CoV-2 test	Virological end point: Proportion of patients with negative COVID-19 PCR test at day 14 in the intention to treat population as per throat swab / change from baseline in viral load	HCQ (600 mg D1-D7) Placebo
8. NCT04403100 (Cardresearch and Cytel, Inc., Brazil)[17]	●	●	●	●	●	214	227	High-risk adults not requiring hospital admission	Nasopharyngeal or saliva sample collection for RT-PCR at days at Screening, days 3, 7, and 14	HCQ 400 mg b.i.d. D1, 200 mg b.i.d. D2-D10 Placebo
9. NCT04308668 (University of Minnesota, USA)[14]	●	●	●	●		244	247	Symptomatic COVID-19 case with confirmed diagnosis within 4 days of symptom onset; OR symptomatic high-risk exposure with known COVID-19 contact and within 4 days of symptom onset	Hospitalizations	HCQ (200 mg tablet; 800 mg orally once, followed in 6 to 8 h by 600 mg, then 600 mg once a day for 4 consecutive days) Placebo Folic acid (USA), lactose (Canada)
10. NCT04334382 (Intermountain Health Care, Inc.; Univ of Utah, USA)		●	●	●		89	88	Outpatients with suspected or confirmed COVID-19	Hospitalizations	HCQ 400 mg p.o. b.i.d. × 1 day, then 200 mg p.o. b.i.d. × 4 days Azithromycin (500 mg p.o. on day 1 plus 250 mg p.o. daily on days 2–5)
11. NCT04329611 (University of Calgary; Alberta Health Services, Canada)[15]	●	●	●	●		111	37	SARS-CoV-2 positive patients within 4 days of first dose of study drug administration	Hospitalizations	HCQ (200 mg po 4 times D1, followed by 200 mg p.o. twice daily D2-D5) Placebo

Abbreviations: COVID-19, coronavirus disease 2019; CQ, chloroquine; D, day; FLS-1, non-hospitalized adult participants with recently confirmed SARS-CoV-2 infection; FLS-2, asymptomatic contacts exposed to a PCR-positive COVID-19 case; HCQ, hydroxychloroquine; PCR, polymerase chain reaction; RT-PCR, real-time polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome-coronavirus 2.

[a]Individuals allocated to the corresponding study arms.

The final study sample for the viral clearance end point (PCR population) included 1560 outpatients with a PCR result for SARS-CoV-2 at baseline and at least one of the follow-up timepoints: 795 participants received HCQ monotherapy, and 765 received an alternative intervention. For the hospitalization end point, 2037 participants were included: 1058 who received HCQ monotherapy and 979 who received a randomized control intervention.

Table 1 summarizes the main characteristics of the study design and interventions of the selected studies. Eight studies were placebo-controlled, two were active-controlled with folic acid, azithromycin, or ascorbic acid, and one compared the effect of HCQ with the standard of care. Eligibility criteria for each study are provided in Table S2 (main criteria) and Table S3 (detailed list of criteria per study) of the supplementary material.

Table S4 summarizes the patient disposition by treatment group, including reasons for discontinuation, for the PCR and viral load populations. The disposition of participants within the HCQ and control groups were balanced in terms of the numbers included in the PCR and viral load analysis populations. Table S5 provides further details regarding the type of data collected for viral clearance in each trial.

Individual participant data integrity

All participants included in the analysis had a positive PCR test for SARS-CoV-2 at study enrollment; however, three studies (NCT04354428, NCT04342169, and NCT04340544) included participants with a negative PCR result at treatment start. A sensitivity analysis was conducted excluding participants with a negative PCR result at treatment start.

There were notable differences in the percentage of SARS-CoV-2 PCR results reported by visit: days 3, 7, 10, and 14. For instance, only two of the eight studies (NCT04354428 and NCT04342169) routinely performed PCR tests on day 10. Studies NCT04304053, NCT04340544, NCT04346667, NCT04351191, NCT04354428, NCT04358068, and NCT04403100 each had the dates of the first and last dose available. The remaining studies did not provide the date of treatment start. A list of missing values for the virological outcome is provided in Table S6. The high number of missing values in cobicistat-boosted darunavir precluded analyses with this arm.

Neither BMI nor weight were available for study NCT0430453 participants, and BMI was not available in study NCT04342169. For NCT0430453, a participant was determined to be low-risk based on age and comorbidities. For study NCT04342169, a participant was determined to be low-risk based on age, comorbidities, and if their weight was less than 91 kg (women) or less than 100 kg (men).

Participant characteristics

Table 2 summarizes the pooled demographic and clinical characteristics of individuals included in both analysis populations. The demographic characteristics were balanced for mean age, sex, race, weight, BMI, global region, and COVID-19 risk were balanced between HCQ and control treatment groups. A broad geographic distribution of participants was represented in this study. Most participants were symptomatic (60%–68%) and most were greater than or equal to 3 days from symptom onset (50–54%) by the time of starting treatment. The HCQ and control groups were balanced regarding days from symptom onset to treatment start.

Primary end points

Figure 2 shows the proportion of participants with a negative, positive or missing SARS-CoV-2 PCR test for each visit. The decline in PCR positive tests in the HCQ and control groups were 22.5% and 22.6% on day 3, 42.0% and 41.6% on day 7, 59.7% and 51.4% on day 10, and 66.7% and 68.2% on day 14. The mixed model longitudinal analysis did not reveal a statistically significant effect of HCQ administration on COVID-19 viral clearance compared to control when participants from all included studies were pooled ($-2LL = 7.66$, $p = 0.18$). The sensitivity analysis removing participants with negative PCR result at baseline showed a similar trend (Table S7).

Table 3 shows the hospitalizations in each group. We found no significant difference in the hospitalization end point between the HCQ and the control group ($-2LL = 0.00$, $p = 0.98$). The percentage of participants who required hospitalization was 3.5% (37/1058) in the HCQ group versus 3.9% (38/979) in the control group (OR 0.995; 95% CI 0.614–1.610). The hospitalization rate among participants who received HCQ was similar, irrespective of the total HCQ dose: 3.4% (2.2–5.0) for low total dose (i.e., ≤3200 mg) and 3.7% (2.2–5.9) for high total dose (i.e., >3200 mg).

Additional analyses

Subgroup analysis for viral clearance

Table S8 summarizes results regarding the primary viral clearance outcome by subgroups of the PCR analysis population: sex, age (<65 years, ≥65 years), risk level (low or high), and the number of days since the first symptoms appeared (categorized as: no baseline symptoms, ≤3 days of symptoms at baseline, >3 days of symptoms at

TABLE 2 Main demographic and clinical characteristics of patients included in the hospitalization and PCR analysis populations

	Hospitalization population		PCR population	
	HCQ ($N = 1058$)	Control ($N = 979$)	HCQ ($N = 795$)	Control ($N = 765$)
Demographic characteristics				
Age (years), mean (SD)	45 (15.60)	46.6 (16.10)	44.3 (16.38)	46.2 (16.74)
Age groups, *n* (%)				
<65 years	948 (89.6)	866 (88.5)	708 (89.1)	675 (88.2)
≥65 years	110 (10.4)	113 (11.5)	87 (25.7)	90 (28.0)
Sex (male), *n* (%)	503 (47.5)	445 (45.5)	351 (44.2)	326 (42.6)
Ethnicity, *n* (%)				
White	256 (24.2)	226 (23.1)	133 (16.7)	95 (12.4)
Mixed	201 (19.0)	202 (20.6)	193 (24.3)	189 (24.7)
Asian	66 (6.2)	34 (3.5)	6 (0.8)	5 (0.7)
Other	57 (5.4)	39 (4.0)	42 (5.3)	27 (3.5)
Missing/not available	442 (41.8)	462 (47.2)	403 (50.7)	437 (57.1)
Region, *n* (%)				
North America	483 (45.7)	373 (38.1)	251 (31.6)	184 (24.1)
South America	210 (19.8)	221 (22.6)	202 (25.4)	210 (27.5)
Europe	246 (23.2)	325 (33.2)	243 (30.6)	319 (41.7)
Asia	119 (11.2)	60 (6.1)	99 (12.5)	52 (6.8)
BMI ≥30 kg/m^2, *n* (%)	228 (33.6)	170 (33.5)	158 (31.9)	125 (32.4)
BMI kg/m^2, mean (SD)	27.9 (5.89)	28.5 (6.14)	27.9 (5.65)	28.5 (5.86)
Comorbidities,[a] *n* (%)				
Hypertension	184 (58.2)	163 (57.2)	136 (62.1)	122 (57.8)
Cardiovascular disease	42 (13.3)	42 (14.7)	35 (16.0)	39 (18.5)
Pulmonary disease	104 (32.9)	87 (30.5)	53 (24.2)	52 (24.6)
Immunocompromised	24 (7.6)	27 (9.5)	21 (9.6)	24 (11.4)
COVID-19 assessments, *n* (%)				
Nasopharyngeal swab performed	459 (78.5)	355 (74.4)	436 (77.9)	337 (74.2)
Oropharyngeal swab performed	118 (20.2)	113 (23.7)	116 (20.7)	109 (24.0)
COVID-19 characteristics, *n* (%)				
COVID-19 symptoms at baseline	697 (65.9)	664 (67.8)	475 (59.7)	475 (62.1)
Days from symptom onset to treatment start, mean (SD)				
≤3 days	133 (12.6)	135 (13.8)	74 (9.3)	67 (8.8)
>3 days	564 (53.3)	529 (54.0)	401 (50.4)	408 (53.3)
Viral load at baseline ($\log_{10}$ copies/ml), mean (SD)	6.4 (2.94)	7.0 (3.37)	6.5 (2.95)	6.9 (3.16)
Risk factor- high, *n* (%)	479 (45.3)	421 (43.0)	338 (42.5)	321 (42.0)
Risk factor- low,[b] *n* (%)	579 (54.7)	558 (57.0)	457 (57.5)	444 (58.0)

Note: Percentages were calculated using the number of participants in each intervention group in the relevant population, with data available, as the denominator.

Abbreviations: BMI, body mass index; COVID-19, coronavirus disease 2019; HCQ, hydroxychloroquine; PCR, polymerase chain reaction.

[a]At least one of the following: pulmonary disease, cardiovascular disease, kidney disease, immunocompromised status, liver diseases, diabetes, and hypertension. Studies NCT04334382, NCT04308668, and NCT04342169 had only weight recorded so BMI was classified using weight thresholds.

[b]Participants indeterminate for risk factor were included in the low-risk group with missing information on age or BMI or comorbidities when low risk for those where information was available.

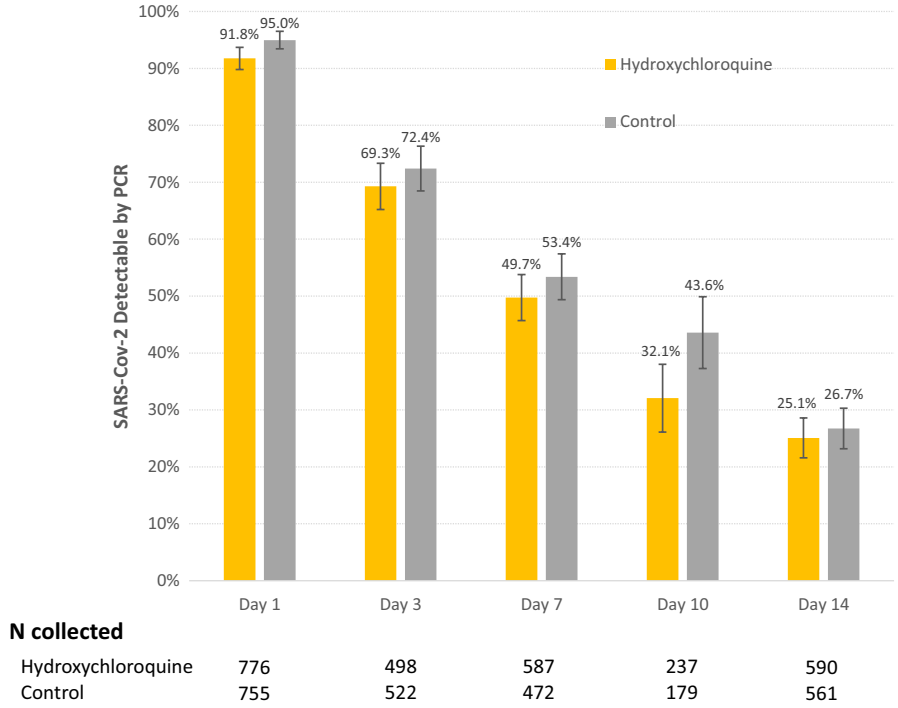

FIGURE 2 SARS-CoV-2 Viral clearance with or without HCQ. Results based on nasal swab samples. Percentage positive are among those samples collected. Samples not collected could be either true missing or not collected per protocol. The mixed model longitudinal analysis did not reveal a statistically significant effect of HCQ administration on viral load clearance over 14 days (−2LL = 7.66, $p = 0.18$). -2LL, log-likelihood ratio test; HCQ, hydroxychloroquine; PCR, polymerase chain reaction; SARS-Cov-2, severe acute respiratory syndrome-coronavirus 2

TABLE 3 Hospitalizations with and without HCQ in randomized trials

Randomized group	Group total *N*	Hospital stay *N*	Proportion hospitalized % (95% CI)	Adjusted probability[a] of hospitalization (95% CI)	Odds ratio[a] (95% CI)	*p* value
HCQ	1058	37	3.5% (2.5, 4.7)	0.0315 (0.0187, 0.0527)	0.995 (0.614, 1.610)	0.98
Control	979	38	3.9% (2.8, 5.2)	0.0317 (0.0183, 0.0541)		

Note: Odds ratio < 1 being in favor of hydroxychloroquine compared with control.

Abbreviations: CI, confidence interval; HCQ, hydroxychloroquine; dose groups are the cumulative total dose.

[a]Probability and odds ratio for hospitalization up to 30 days are estimates obtained from logistic linear mixed model that accounts for study-specific random effects and includes fixed effects of treatment, risk factor and days since onset of symptoms variables.

baseline). The same analyses, excluding participants with negative PCR at baseline, are summarized in Table S9.

Significant differences regarding the percentage of participants with viral clearance at days 3 and 7 were detected only in participants in the high-risk subgroup ($p = 0.04$; Table S10). However, after removing the participants with negative PCR results at baseline, the significant difference in this high-risk subgroup was no longer detected ($p = 0.66$; Table S9). Of note, the percentage of negative PCR results at baseline was unbalanced in the high-risk subgroup: 11% in HCQ versus 5% in the control group.

There was no observable weight-related dosing effect with HCQ, and no observable difference in clearance of PCR positivity by weight in either group (Table S10, pooled studies NCT04342169 and NCT04403100). There was no effect of HCQ by weight, on the clearance of virus by nasal swab on day 3. Table S11 summarizes the weight-related dosing effect for each of the two studies reported separately.

Viral load

Data to assess the changes in viral load over time were compiled from two studies: NCT04304053 with the two separate cohorts (FLS-1 and FLS-2), and NCT04340544. The two studies assessed this parameter on (days 1, 3, 7, 10, and 14). There were no differences observed in mean viral load rates between HCQ group vs control (Table S12) at any timepoint.

DISCUSSION

Our meta-analysis of IPD provides evidence for cessation of HCQ as a therapeutic approach for outpatients with mild COVID-19. HCQ administration had no effect on the proportion of trial participants with negative PCR for SARS-CoV-2 at any of the time points assessed (i.e., from 1 to 14 days). On day 10 after treatment start, a higher proportion of individuals had a negative result, although only

two studies reported day 10 PCR test results (missingness >70%). The analysis of hospitalization risk did not reveal significant differences between controls and HCQ-treated participants, irrespective of the dose received.

The lack of efficacy of HCQ for preventing COVID-19 infection among exposed contacts was well-established in two large RCTs conducted early after the COVID-19 become a pandemic.[19,20] Likewise, failure of HCQ to improve outcomes in hospitalized patients with severe/critical COVID-19 was observed in the RECOVERY and WHO Solidarity trials, both of which discontinued HCQ arms due to lack of effect of therapy in this setting.[21,22] These findings were confirmed in a meta-analysis of trials recruiting hospitalized patients, which showed no significant effect with low heterogeneity.[12] However, research in the mild COVID-19 outpatient setting has been limited for two reasons. First, most trials did not reach the expected sample size. Second, the heterogeneity of different trials on the dose, dose interval, and populations among different trials either requires the exclusion of many individual trials in conventional meta-analyses or renders the veracity of the estimates uncertain. Moreover, the validity of aggregate data meta-analyses is affected by the reporting quality of the RCTs and inconsistent definition of outcomes across included trials.[23–27]

Unlike conventional meta-analyses, which rely on aggregate data of trials addressing the same research question, in this IPD meta-analyses, we collected both published and unpublished data from eligible primary studies, derived standardized outcome definitions, used a consistent unit of analysis across included RCTs, and assessed interactions between interventions and participants' characteristics.[28] This approach allowed us to analyze the hospitalization risk on a dataset of 2037 outpatients (1058 treated with HCQ) with confirmed COVID-19.

Regarding the heterogeneity of viral clearance outcomes, most trials used a consistent criterion for the event (i.e., negative result in a SARS-CoV-2 PCR test result), but the time frame for the assessment varied substantially among trials. In meta-analyses of aggregated data, this heterogeneity requires separate analyses in which only trials with matching end points can be analyzed together, which often makes the meta-analysis impossible despite the inclusion of studies assessing viral clearance.[24,26] Conversely, the individual participant data approach allowed matching individual patients who were tested at the same timepoint.

Our analysis has some limitations. First, IPD meta-analyses are limited to investigators who are willing to share their data. It is more difficult and time-consuming for RCT investigators to conduct an IPD meta-analysis (e.g., establishing data sharing agreements) and therefore selection bias could affect validity of results. Second, despite using a comprehensive literature search strategy and establishing an a priori protocol, we did not obtain IPD from all trials identified because some of them were still recruiting. We had to balance the need to include as many studies as possible while also being expedient in initiating the analyses. On the other hand, a key strength of this study is that we established a data analysis plan, checked data integrity, and clarified uncertainties with individual researchers when needed.

In summary, although HCQ is no longer a research priority, there is no convincing evidence on the efficacy of HCQ in the treatment of outpatients with mild COVID-19. Our meta-analysis provides evidence to support the interruption in the use of HCQ in mild COVID-19 outpatients to reduce progression to severe disease.

AUTHOR CONTRIBUTIONS

O.M., M.C., D.C., and M.D. wrote the manuscript. O.M., G.R., D.R.B., A.M.S., A.S., C.J., B.W., M.D.H., D.S., P.K., and M.D. designed the research. O.M., G.R., D.R.B., A.M.S., A.S., C.J., B.W., M.D.H., D.S., P.K., M.C., T.C.L., K.H.H., E.G.M., R.H., M.H., J.M.B., I.S., L.M., L.R., K.C., E.D., and D.W. performed the research. D.C. and M.C. analyzed the data.

ACKNOWLEDGMENTS

The authors would like to thank Elnaz Menhaji-Klotz, John Eikelboom, Satyapal Ingle, Jintanat Ananworanich, Robin Mogg, Maria Kudela, Adriana Andrate, Arzhang Cyrus Javan, Elizabeth Church, and Nathalie Strub-Wourgaft for their contribution to the project. We also thank Debbie Daehnick and Gerard Carot-Sans for providing medical writing support with manuscript preparation.

FUNDING INFORMATION

This study was funded by Grifols Worldwide Operations Ltd. (Dublin, Ireland), the Crowdfunding campaign YoMeCorono (https://www.yomecorono.com/ca/), COVID Therapeutics Accelerator, Bill & Melinda Gates Foundation and the Bill & Melinda Gates Medical Research Institute.

CONFLICT OF INTEREST

C.J. has been consultant to AbbVie, Gilead, received royalties from UpToDate, and participated in a DSMB for Medpace. D.S. has received honoraria or fees from Bayer Pharmaceuticals, Linear Therapies, Matrix BioMed, Fluxergy, Kiadis, Model Medicine, Signant Health, and VxBiosciences. All other authors declared no competing interests for this work.

ORCID

Marc Corbacho https://orcid.org/0000-0001-8675-2868
Rachel Hess https://orcid.org/0000-0003-2545-8504

REFERENCES

1. Dong L, Hu S, Gao J. Discovering drugs to treat coronavirus disease 2019 (COVID-19). *Drug Discov Ther*. 2020;14:58-60.
2. De Duve C. Lysosomes revisited. *Eur J Biochem*. 1983;137:391-397.
3. Vincent MJ, Bergeron E, Benjannet S, et al. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virol J*. 2005;2:1-10.
4. Savarino A, Di Trani L, Donatelli I, Cauda R, Cassone A. New insights into the antiviral effects of chloroquine. *Lancet Infect Dis*. 2006;6:67-69.
5. 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.
6. 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.
7. 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*. 2020;71:732-739.
8. Dutta D, Sharma M, Sharma R. Short-term hydroxychloroquine in COVID-19 infection in people with or without metabolic syndrome–clearing safety issues and good clinical practice. *Eur Endocrinol*. 2020;16:109-112.
9. Marin S, Martin Val A, Bosch Peligero M, et al. Safety of short-term treatments with Oral chloroquine and hydroxychloroquine in patients with and without COVID-19: a systematic review. *Pharmaceuticals*. 2022;15:634.
10. Oren O, Yang EH, Gluckman TJ, Michos ED, Blumenthal RS, Gersh BJ. Use of chloroquine and hydroxychloroquine in COVID-19 and cardiovascular implications: understanding safety discrepancies to improve interpretation and design of clinical trials. *Circ Arrhythm Electrophysiol*. 2020;13:e008688.
11. Geleris J, Sun Y, Platt J, et al. Observational study of hydroxychloroquine in hospitalized patients with Covid-19. *N Engl J Med*. 2020;382:2411-2418.
12. Axfors C, Schmitt AM, Janiaud P, et al. Mortality outcomes with hydroxychloroquine and chloroquine in COVID-19 from an international collaborative meta-analysis of randomized trials. *Nat Commun*. 2021;12:1-13.
13. Self WH, Semler MW, Leither LM, et al. Effect of hydroxychloroquine on clinical status at 14 days in hospitalized patients with COVID-19: a randomized clinical trial. *JAMA*. 2020;324:2165-2176.
14. Skipper CP, Pastick KA, Engen NW, et al. Hydroxychloroquine in nonhospitalized adults with early covid-19: a randomized trial. *Ann Intern Med*. 2020;173:623-631.
15. Schwartz I, Boesen ME, Cerchiaro G, et al. Assessing the efficacy and safety of hydroxychloroquine as outpatient treatment of COVID-19: a randomized controlled trial. *C Open*. 2021;9:E693-E702.
16. Johnston C, Brown ER, Stewart J, et al. Hydroxychloroquine with or without azithromycin for treatment of early SARS-CoV-2 infection among high-risk outpatient adults: a randomized clinical trial. *EClinicalMedicine*. 2021;33:100773.
17. Reis G, Moreira Silva EAS, Medeiros Silva DC, et al. Effect of early treatment with hydroxychloroquine or lopinavir and ritonavir on risk of hospitalization among patients with COVID-19: the TOGETHER randomized clinical trial. *JAMA Netw Open*. 2021;4:e216468.
18. Mitjà O, Corbacho-Monné M, Ubals M, et al. Hydroxychloroquine for early treatment of adults with mild Covid-19: a randomized-controlled trial. *Clin Infect Dis*. 2020;73:e4073-e4081. doi:10.1093/cid/ciaa1009
19. Boulware DR, Pullen MF, Bangdiwala AS, et al. A randomized trial of hydroxychloroquine as postexposure prophylaxis for Covid-19. *N Engl J Med*. 2020;383:517-525.
20. Mitjà O, Corbacho-Monné M, Ubals M, et al. A cluster-randomized trial of hydroxychloroquine for prevention of Covid-19. *N Engl J Med*. 2021;384:417-427.
21. The RECOVERY Collaborative Group. Effect of hydroxychloroquine in hospitalized patients with Covid-19. *N Engl J Med*. 2020;383:2030-2040.
22. World Health Organization (WHO). WHO discontinues hydroxychloroquine and lopinavir/ritonavir treatment arms for COVID-19. *Dent News*. 2020. Accessed December 31, 2022. https://www.who.int/news/item/04-07-2020-who-discontinues-hydroxychloroquine-and-lopinavir-ritonavir-treatment-arms-for-covid-19
23. Tanni S, Bacha HA, Naime A, Bernardo WM. Use of hydroxychloroquine to prevent SARS-CoV-2 infection and treat mild COVID-19: a systematic review and meta-analysis. *J Bras Pneumol*. 2021;47:e20210236.
24. Kumar J, Jain S, Meena J, Yadav A. Efficacy and safety of hydroxychloroquine/chloroquine against SARS-CoV-2 infection: a systematic review and meta-analysis. *J Infect Chemother*. 2021;27:882-889.
25. Singh B, Ryan H, Kredo T, Chaplin M, Fletcher T. Chloroquine or hydroxychloroquine for prevention and treatment of COVID-19. *Cochrane Database Syst Rev*. 2021;2021: CD01358.
26. Kashour Z, Riaz M, Garbati MA, et al. Efficacy of chloroquine or hydroxychloroquine in COVID-19 patients: a systematic review and meta-analysis. *J Antimicrob Chemother*. 2021;76:30-42.
27. Martins-Filho PR, Ferreira LC, Heimfarth L, Araújo AAdS, Quintans-Júnior LJ. Efficacy and safety of hydroxychloroquine as pre-and post-exposure prophylaxis and treatment of COVID-19: a systematic review and meta-analysis of blinded, placebo-controlled, randomized clinical trials. *Lancet Reg Heal Am*. 2021;2:100062.
28. Stewart LA, Clarke M, Rovers M, et al. Preferred reporting items for a systematic review and meta-analysis of individual participant data: the PRISMA-IPD statement. *JAMA*. 2015;313:1657-1665.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Mitjà O, Reis G, Boulware DR, et al. Hydroxychloroquine for treatment of non-hospitalized adults with COVID-19: A meta-analysis of individual participant data of randomized trials. *Clin Transl Sci*. 2023;00:1-12. doi:10.1111/cts.13468