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Letter to Editor

Chloroquine/Hydroxychloroquine and SARS-CoV 2; Lessons from Case-Control Studies

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Dear Editor. In December 2019, a novel viral pneumonia caused by Severe Acute Respiratory Syndrome Coronavirus type 2 (SARS-CoV 2) was emerged in Wuhan, Hubei Province, China [1]. We are lives in global pandemic of coronavirus disease 2019 (COVID-19); However, we have limited therapeutic agents for treatment of COVID-19 and reduction of mortality caused by this virus; there is no FDA approved option against SARS-CoV 2 which is global concern [2]. According to review of the literatures, there are suggested several drugs including Remdesivir, Favipiravir, Ribavirin, Interferons, Lopinavir/ritonavir, Oseltamivir, Chloroquine, hydroxychloroquine, and azithromycin, Convalescent plasma, Herbal medications, non-steroidal antiinflammatory drugs (NSAIDs), Mycophenolic acid, Monoclonal or polyclonal antibodies, and, Angiotensin-converting enzyme 2 gene-based peptides for combating with SARS-CoV 2 [3]; but there is no conclusive evidence for efficacy and safety of these therapeutic agents against COVID-19 [4-5].

The Lopinavir/ritonavir was initially recommended for treatment of COVID-19; but Cao et al., 2020 showed that there is no clinical benefit in COVID-19 cases which are received lopinavir-ritonavir and control group [5-6]. Subsequently, it's suggested that Chloroquine/Hydroxychloroquine can be used against SARS-CoV 2 (2-4).

Chloroquine (CQ) has several benefits such inhibitory effects in SARS-CoV-2 infected Vero-E6 cell lines (EC50 = 1.13 μM), increasing endosomal pH, dysregulating the glycosylation of angiotensin-converting enzyme 2 receptors, immunomodulatory activity, provoke T regulatory cells, as well as, clinically beneficial effect in recently clinical trials which is conducted by Huang et al., 2020 (2-3,6). Moreover, there is concern about CQ due to its toxicity and Hydroxychloroquine (HCQ) was introduced as alternative option of CQ; particularly, HCQ has inhibitory roles in SARS-CoV infected Vero cells (0.72 μM) [7]. Therefore, CQ/HCQ has emerged as potential therapeutic option against SARS-CoV 2; there are numerous registered clinical trial which are evaluating the clinical benefit of CQ/HCQ for treatment of COVID-19 .

Herein, the aim of study was evaluation of clinically benefits of CQ/HCQ for treatment of COVID-19 using statistical analysis of provided evidence. We conducted a systematic search in several databases including PubMed, Scopus, Embase, EBSCO, Google scholar, Cochrane library, medRxiv, and bioRxiv to retrieving all available case-control articles in relation to efficacy of CQ/HCQ in treatment of SARS-CoV 2. we have no limit in language or date in searching and data collection; also, we used from several keywords according to MeSH including "2019-nCoV", "2019 novel coronavirus", "COVID-19", "coronavirus disease 2019", "chloroquine",

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"hydroxychloroquine", and "Plaquenil" for searching databases. In the next, titles, abstracts of all obtained studies were screened to remove duplicates and collected all relevant case-control studies. Subsequently, the full-text of relevant studies were evaluated carefully and required data were extracted in the Table 1. Finally, clinical benefit of CQ/HCQ was measured with odds ratio with 95% confidence intervals.

Table 1. Characteristics of included studies

Study	Study type	Country	Age	No. of patients		Severity of disease	HCQ /CQ dose	Improvement		Viral clearance		Death	Severity progression			Ref
				Case	Control			Case	Control	Case	Control	Case	Control	Case	Control	
Gautret	nRCT	France	45.1	20	16	Moderate	600 mg/d	NR	NR	14	2	NR	NR	NR	NR	8
Mahevas	Retrospective	France	60	84	97	Moderate/Pneumonia requiring O2	600 mg/d	NR	NR	NR	NR	2.8%	4.6%	24	23	9
Mallat	prospective	UAE	37	23	11	Moderate/Pneumonia requiring O2	400 mg/d	NR	NR	47.8%	90.9%	NR	NR	NR	NR	10
Tang	RCT	China	46	75	75	Moderate	1200 mg/d	59.9%	66.6%	85.4%	81.3%	NR	NR	NR	NR	11
Jun	RCT	China	30	15	15	Moderate	400 mg/d	15	15	13	14	NR	NR	5	7	12
Chen	RCT	China	44.7	31	31	Moderate	400 mg/d	25	17	NR	NR	NR	NR	0	4	13
Rosenberg	Retrospective	USA	NR	271	431	Moderate	400 mg/d	NR	NR	NR	NR	54/271	21/211	52	50	14
Huang	RCT	China	44	10	12	Moderate	400 mg/d	60%	25%	10	12	NR	NR	NR	NR	6
Geleris	RCT	USA	NR	811	565	Mild/Moderate	400 mg/d	NR	NR	65.0%	18.8%	262	84	NR	NR	15
Huang	Prospective	China	43	197	176	Mild/Sever	500 mg/d	NR	NR	95.9%	79.6%	NR	NR	NR	NR	16
Barbosa	qRCT	USA	62.7	32	31	Moderate	800 mg/d	NR	NR	NR	NR	6	2	NR	NR	17
Magagnoli	Retrospective	USA	68	198	395	Moderate	NR	124	255	NR	NR	38	37	NR	NR	18
Membrillo	Retrospective	Spain	51.5	123	43	Mild/Sever	800 mg/d	70	19	NR	NR	27	21	NR	NR	19
Yu	Retrospective	China	68	48	502	Mild/Sever	400 mg/d	NR	NR	NR	NR	9	238	NR	NR	20

We analyzed the efficacy of CQ/HCQ in 1) clinically improvement of COVID-19 patients, 2) SARS-CoV 2 clearance (by PCR), 3) reduction of death, and 4) prevention form progression of disease severity. All statistical analysis was conducted using Comprehensive Meta-Analysis (CMA) software version 2.2 (Biostat, Englewood, NJ, USA). The pooled analysis was measured by fixed-effect models; However, in high heterogeneity cases, I2 index>25% and Cochrane Q test p-value ≤0.05, we used from random effect model based on Dersimonian and Laried method. In addition, publication bias was assessed by Eggers p-Value and Beggs p-Value .

We provided 14 relevant case-control studies (total participate: 4,338) in regarding to evaluation of CQ/HCQ beneficial effects in treatment of COVID-19. The study types were RCT, nRCT, qRCT, retrospective, and prospective. In the present report, there is 1,938 SARS-CoV 2 infected cases which received CQ/HCQ as well as 2,400 patients as control (not received any dose of CQ/HCQ). The mean age of patients was estimated about 50.2 years. The patient status was mild to severe requiring O2. This studies were conducted in France, China, USA, Spain, and UAE.

According to statistical analysis, there is no significant benefit in clinical improvement of COVID-19 patients were received CQ/HCQ in comparison with control (OR:1.089; 0.82-1.432 with 95% CIs; p-Value: 0.54; I2: 58.97; Q-Value: 9.75; p-Value: 0.045; Eggers p-Value: 0.51; Beggs p-Value: 0.04).

In addition, CQ/HCQ administration has not preventive effects on mortality due to COVID-19 (OR: 2.03; 1.66-2.47; p-Value: 0.001; I2: 91.86; Q-Value: 73.72; p-Value: 0.001; Egers p-Value: 0.13; Beggs p-Value: 0.18). However; there is significant increasing of viral clearance in CQ/HCQ treated cases (OR:6.50; 5.17-8.19; p-Value: 0.001; I2: 85.74; Q-Value: 35.0; p-Value: 0.001; Eggers p-Value: 0.06; Beggs p-Value: 0.12). Therefore, Although CQ/HCQ was efficient in SARS-CoV 2 virological cure; but there is no efficient in recovery and improvement of patients. Also, CQ/HCQ has no preventive effect on mortality of COVID-19 patients (Hazard Ratio: 1.073; 0.89-1.29 with 95% CIs; p-Value: 0.46; I2: 72.15; Q-Value: 14.36; p-Value: 0.006; Eggers p-Value: 0.38; Beggs p-Value: 0.50). Unfortunately, we found that disease severity (transfer to ICU; radiological chest progression, require to mechanical ventilation, and develop into acute respiratory distress syndrome) was significantly higher among COVID-19 patients were received CQ/HCQ which is represent inefficacy of these drugs in treatment of COVID-19 (OR:

1.49; 1.05-2.10; p-Value: 0.023; I2: 48.92; Q-Value: 5.87; p-Value: 0.11; Eggers p-Value: 0.001; Beggs p-Value: 0.044) .

Chloroquine has several disadvantages consisting hypoglycemia, diarrhea, prolong QTc interval, and AV block [21-22]. In addition, Gendelman et al., recently published an article about inefficacy of continuous Hydroxychloroquine or colchicine therapy for prevention of SARS-CoV 2 infection [23]. According to recent RCTs, HCQ has no therapeutic effects on prevention or improvment of major outcomes in COVID-19 patients [24-25]. According to current analysis, it seems that CQ/HCQ has not benefit in treatment of COVID-19 patients. But we have several limitations such as 1) high heterogeneity rate which reduce reliability of analysis, 2) presence of publication bias, 3) dosage of CQ/HCQ was varying among the included studies, 4) different outcome between studies, 5) various in times of evaluating outcomes, and 6) low sample size. We need to complete further clinical trials to confirmed our results.

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Conflict of interest

The authors have no conflict of interest.

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