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Acute effects of hydroxychloroquine prophylaxis for COVID-19 in health care professionals – An online survey

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Hydroxychloroquine, an antimalarial, is being used worldwide for prophylaxis and treatment of Corona virus disease-19 (COVID-19). Though the drug is commonly used in many chronic inflammatory diseases for protracted periods, its safety in the new indication is still under scrutiny. Therefore, this institute based study sought to assess the acute adverse effects of hydroxychloroquine among in-house health care professionals who were taking the drug for COVID-19 prophylaxis. A questionnaire seeking information on the use of the drug was prepared and disseminated electronically to the target population. The responses were also received electronically and analysed. The participants (n=54) had taken prophylaxis for 1-7 weeks. The most common adverse effects in the cohort were nausea (02) and skin rash (02). The total number of adverse effects reported by the participants was 08. One incidence each of gastric upset (01), dizziness (01), pain abdomen (01), and chest tightness (01) was reported. None of the adverse effects were serious. Our study indicates that the prophylactic weekly single dose of hydroxychloroquine is not associated with any serious adverse effects within 1-7 weeks of initiation. Elucidation of the long term and chronic adverse effects, if any, requires further studies.

Keywords: Hydroxychloroquine, Ondansetron, Prophylaxis, Systemic lupus erythematous

Coronaviruses are a large family of viruses that cause illness in humans and animals. In humans, several corona viruses cause respiratory infections which can present as mild to more severe diseases, such as Middle East Respiratory Syndrome (MERS) and Severe Acute Respiratory Syndrome (SARS)[1,2].

In the month of December 2019, Wuhan, China first reported that patients were suffering from an infection characterized with atypical pneumonia, fever, dry cough, and progressive dyspnoea with a novel Beta coronavirus named 2019 novel coronavirus (2019-nCoV)[1,3]. On 11th February 2020, the virus and the disease were renamed as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and COVID-19 by the International Committee on Taxonomy of Viruses (ICTV) and World Health Organization (WHO), respectively[4,5]. Within around three months, the viral infection spread rapidly to most parts of the world, prompting the WHO to declare COVID-19 as pandemic on 11th March 2020. In absence of any effective drug against the virus, the infection rate and mortality are high and as per the information with the WHO-COVID-19 dashboard, as of 31st July 2020, there have been 17, 106, 007 confirmed cases of COVID-19, including 668, 910 deaths worldwide[4]. Any hope of controlling the pandemic with a newly developed drug is far from practical because drug development is a time-taking and costly process. Testing drugs from the existing "armamentarium" of approved drugs (but for other indication) against a disease, popularly known as drug repurposing, is a viable alternative in the current scenario. Drugs like hydroxychloroquine (HCQ),

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Abbreviations: 2019-nCov, 2019-novel coronavirus; ACE2, Angiotensin converting enzyme 2; AZT, Azithromycin; CQ, Chloroquine; COVID-19, Corona virus disease-19; Cgas, Cyclic GMP/AMP synthase; ER, Endoplasmic reticulum; HCQ, Hydroxychloroquine; ICMR, Indian Council of Medical Research; ICTV, International Committee on Taxonomy of Viruses; IVM, Ivermectin; MERS, Middle East respiratory syndrome; PTM, Post-translational modification; RT-PCR, Real time- polymerase chain reaction; SARS, Severe acute respiratory syndrome; SARS-Cov-2, Severe acute respiratory syndrome coronavirus-2; STING, Stimulation of interferon genes; SLE, Systemic lupus erythematosus; TLR, Toll-like receptors; TGA, Trans-Golgi apparatus; WHO, World Health Organization.

ivermectin (IVM), and azithromycin (AZT) have potential efficacy against the virus and are already in clinical trials worldwide[2]. HCQ and IVM are known to act by creating an acidic environment and inhibiting the importin (IMPα/β1) mediated viral import, respectively. Like HCQ, AZT (a weak base) is observed to have an acidotropic action. The anti-viral action of these drugs including chloroquine (CQ) and its congener HCQ were noted *in vitro* which warranted further exploration in the infection[6]. While CQ is better known for the treatment of malaria, HCQ is commonly used for the treatment of chronic diseases like rheumatoid arthritis and Systemic lupus erythematosus (SLE)[7]. HCQ is preferred in these chronic conditions because of lesser toxicity and long-term side effects compared to CQ. HCQ is synthesised by the addition of a hydroxyl group at the end of the side chain of CQ (Fig. 1). This minor structural modification alters the pharmacokinetics of the molecule, such that the molecule has decreased tissue affinity and lesser accumulation in tissues, and therefore, lesser chances of retinal damage and corneal opacity when prescribed for longer periods. In addition to the better safety profile of HCQ, the efficacy of antiviral activity of the molecule has also been found superior to CQ in *in vitro* studies[6].

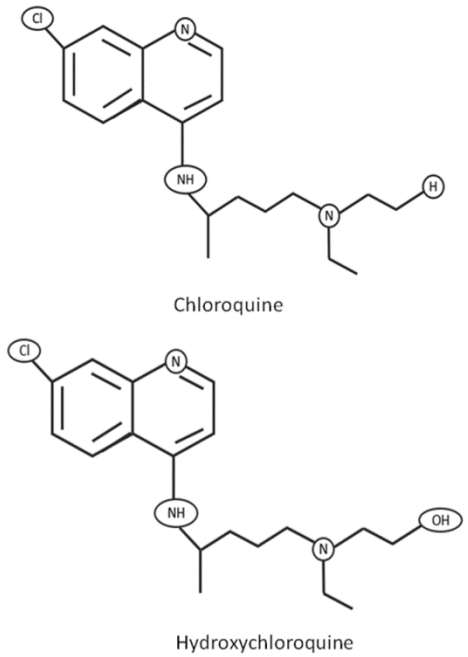

Fig. 1 — Structure of chloroquine and hydroxychloroquine. Chemically, chloroquine is 4-*N*-(7-chloroquinolin-4-yl)-1-*N*, 1-*N*-diethylpentane-1,4-diamine. Substitution of the H atom at the end of diethyl-pentane side chain with hydroxyl moiety yields hydroxychloroquine

On 23rd March 2020, the Indian Council of Medical Research (ICMR) recommended hydroxychloroquine as prophylaxis to asymptomatic healthcare workers involved in the care of suspected or confirmed cases of COVID-19 and asymptomatic household contacts of laboratory-confirmed cases with the dose of 400 mg twice daily in day one, then 400 mg once weekly for 7 weeks for healthcare providers and 3 weeks for asymptomatic household contacts. The prophylactic therapy excludes the children below 15 years of age[8]. Thereafter, on the 22nd May 2020 ICMR recommended to continuing weekly prophylactic therapy for health care providers (involved in COVID-19 related activities) with strict monitoring of clinical and ECG parameters[9].

Aims and Objectives

In this study, we sought to study the safety and tolerability of the initiation of Hydroxychloroquine in the prophylaxis of COVID-19 in a cohort of health care professionals.

Subjects and Methods

This study is limited to a tertiary care centre in eastern India directly involved in the management of COVID-19 patients. A 100 bedded covid dedicated hospital, an RT-PCR testing laboratory for COVID-19, a separate fever clinic, and a quarantine unit with health care personnel, managed and run by health care providers of the institution. According to the advisory recommendation of ICMR, prophylactic therapy of HCQ is being voluntarily taken by many health care professionals. This web- based study was conducted to collect data on the potential adverse effects of prophylactic use of HCQ for COVID-19 by health care professionals of the institution.

Subject Selection Criteria

Healthcare providers who were taking HCQ as prophylaxis for COVID-19 were included in this study. Those persons taking HCQ due to autoimmune diseases like rheumatoid arthritis or Systemic Lupus Erythematous (SLE) or any other conditions were excluded from the study.

Study Tool

A questionnaire was prepared using a set of questions to elicit information on demographic, clinical characteristics, drug-taking behaviour, and adverse effects of HCQ in the respondents. The questionnaire was transcribed in Google Form (a web-

based survey tool) and was disseminated electronically using freely available mobile phone applications. Health care providers were requested to fill up and submit the form as per convenience. No force or coercion was employed during the study and the employees were free to take decision to participate in the study. The study and the questionnaire were approved by the Institutional Ethics Committee (IEC) (Ref. No. F-24/PR/COMJNMH/IEC/20/318)

Statistical Analysis

Data were processed and analysed at the Department of Pharmacology, College of Medicine and JNM Hospital, WBUHS, Kalyani. Analyses were done only after the last participant submitted the response on 31st May 2020. Frequency in groups and the number of events were reported. Percentage calculation was used wherever applicable.

Results and Analysis

Fifty-four (32 males and 22 females) health care personnel of the institution filled-up and submitted the questionnaire, during April-May 2020. The data in all the submissions were complete and eligible for analysis. The number of participants in the age (in years) range <30, 30-39, 40-49, 50-59, and >60 was 3, 26, 18, 6 and 1, respectively. While the majority of the participants were medical doctors (post-graduates/graduates=46/4), 4 non-doctor (post-graduates/graduates=2/2) participants were present in the final sample size of 54. Most of the participants were on prophylactic HCQ even before the commencement of the study. The number of participants who had completed 1, 2, 3, 4, 5, 6, and 7 weeks of therapy at the time of submission of the form was 2, 2, 7, 21, 11, 6, and 5, respectively. Although ICMR advisory is to take HCQ with a meal, the majority of participants (43) took HCQ after meal. Only 6 participants (11. 11%) accurately followed the guideline of consuming the medicine during the meal. A few participants (5) even consumed the drug before meal. The incidental and pre-existing presence of hypertension, diabetes, asthma, peptic ulcer/gastritis and ankylosing spondylosis was observed in 8, 2, 6, 1, and 1 participants, respectively. The remaining 36 subjects were free from any co-morbid condition. The total number of adverse effects reported by the participants following prophylactic HCQ was 08. Two participants experienced nausea which was reportedly relieved by oral self-administration of the anti-emetic ondansetron in the recommended dose. The adverse effect in both the participants had a temporal association with untimely intake of the drug with respect to meals- one had taken before a meal and the other had taken the medicine after light breakfast. A skin rash was also reported by 2 persons. Chest tightness was seen in one case, which however, recovered after 24 h without any medication. This chest tightness was experienced on repeat dose and the person stopped the use of hydroxychloroquine after 2nd week. One incidence each of gastric upset (01), dizziness (01), and pain abdomen (01) was also reported. All the adverse effects were observed in the healthy participants and those with pre-existing ailments did not report any adverse event. No relation to the age of the participants was seen.

Discussion

Though the molecular action of hydroxychloroquine in COVID-19 is not known, several mechanisms have been postulated. The inhibitory action of the drug starts even before the virus could gain entry inside the cells. The virus interacts with angiotensin- converting enzyme 2 (ACE2) on the host cell surface for internalisation. ACE2 is transmembrane glycoproteins with terminal sialic acid residues which act as the precise sites of the virus-host cell surface interaction. Hydroxychloroquine inhibits the cell membrane enzyme quinone reductase 2 in the sialic acid biosynthetic pathway (Vide No. 1 in Fig. 2). The drug, therefore, causes failure of glycosylation of ACE2, leading to diminished virus-host receptor interaction. The entry of the virus into the host cell for propagation is curtailed and the chances of infection and progression of infection are reduced[10,11]. The inhibitory action of the drug against the virus continues inside the cell. Once inside the cell, the viral envelope fuses with the endosomal membrane and its nucleic acid is released into the cytoplasm (Vide No. 2 in Fig. 2). This step is the trigger for viral replication and is heavily dependent on the acidic pH of the endosomal content. Chloroquine or its hydroxylated derivative accumulates inside and alkalinise the endosomal milieu and impedes the release of the viral genome[12]. After the replication phase, there is a post-translational modification (PTM) of the viral proteins in the acidic milieu of the endoplasmic reticulum (ER) and trans-Golgi apparatus (TGA) (Vide No. 3 in Fig. 2). Finally, the assembly of replicated nucleic acid and translated viral proteins occurs, leading to the shedding of a huge number of viral progeny (Fig. 2). The drug, similar to its action in the endosomes, elevates the pH in ER and TGA and interferes with the step of PTM of the viral

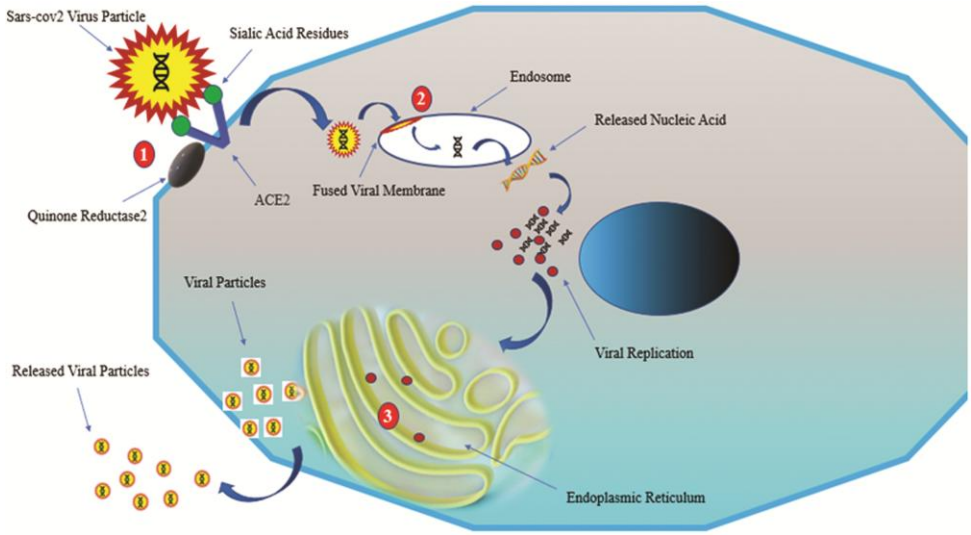

Fig. 2 — Anti-viral activity of hydroxychloroquine (HCQ) against SARS-CoV-2

proteins[13]. The actions of the drug constitute a "triune trouble" for the virus whereby the virus is chronologically inhibited at the three stages of the viral life cycle-entry, replication, and post-translational modification.

Immuno-modulatory actions of the drug against the virus have been identified at three cellular sites-lysosomes, endosomes and the cytoplasm. In the two organelles,the drug shifts the pH from acidic to basic. Such alterations in the lysosomes of the antigen-presenting cells (B-cells, Dendritic cells), prevent the processing and presentation of antigens to the T-cells, impeding their activation. The production of inflammatory cytokines (IL-1, IL-6, TNF-α *etc.*) by both the T and B cells are reduced (Vide No. 1 in Fig. 3). Similarly, altered pH in the endosomes interferes with binding between toll-like receptors (TLR7 and TLR9), and their RNA/DNA ligands and the inflammatory TLR signalling is suppressed[11] (Vide No. 2 in Fig. 3).The anti-inflammatory action of the drug in the cytoplasm is by the prevention of the cytosolic DNA-cyclic GMP/AMP synthase (cGAS) interaction. The interaction is known to increase inflammation by the Stimulation of Interferon Genes (the STING pathway) (Vide No. 3 in Fig. 3). So, the action mediated by the drug at the three sites-endosomes, lysosomes, and cytoplasm lead to precise anti-inflammatory action. The suppression of cytokine production can be life-saving because it is believed that the potentially fatal cytokine storm syndrome occurs due to the prodigious release of the immune mediators[14].

The *in vitro* anti-viral inhibitory actions of hydroxychloroquine together with the elicitation of its molecular mechanism of action has prompted many agencies to recommend its use for both treatment and prophylaxis. Being in use for chronic inflammatory conditions, the assumed safety of the drug and easy availability has lead to widespread use in the ongoing pandemic. Within a short period of time, conflicting reports on safety and efficacy have flooded the existing database of evidence, leaving much of the fraternity divided on its future use. At this juncture, it becomes imperative to investigate the role of the drug further. While the gold standard of efficacy analysis is a randomized controlled trial, which is being actively conducted globally, the issue of safety and tolerability can be addressed with the meticulously designed observational study. In the pandemic situation, where social distancing is the necessity rather than the norm, we conducted the entire study over the web. Though "armed" with a humble sample size of 54 "front line warriors", the results are nevertheless optimistic as far as safety is concerned. In our study, hydroxychloroquine induced few adverse effects (8 episodes in 54 subjects); all were mild in nature. No serious adverse effect was seen throughout this period. Out of 54 persons, 2 persons had the experience of nausea, relieved by a single dose of oral ondansetron of 4 mg. Out of these 2 participants, one person took hydroxychloroquine before a meal and another person received after light breakfast. ICMR advised to take hydroxychloroquine during the meal to avoid this type of adverse effect. Skin rash was also noted in 2 persons. One adverse effect of chest tightness was reported. The tightness disappeared after 24 h without any medication. The drug was discontinued after a similar episode was seen with the drug in the 2nd week.

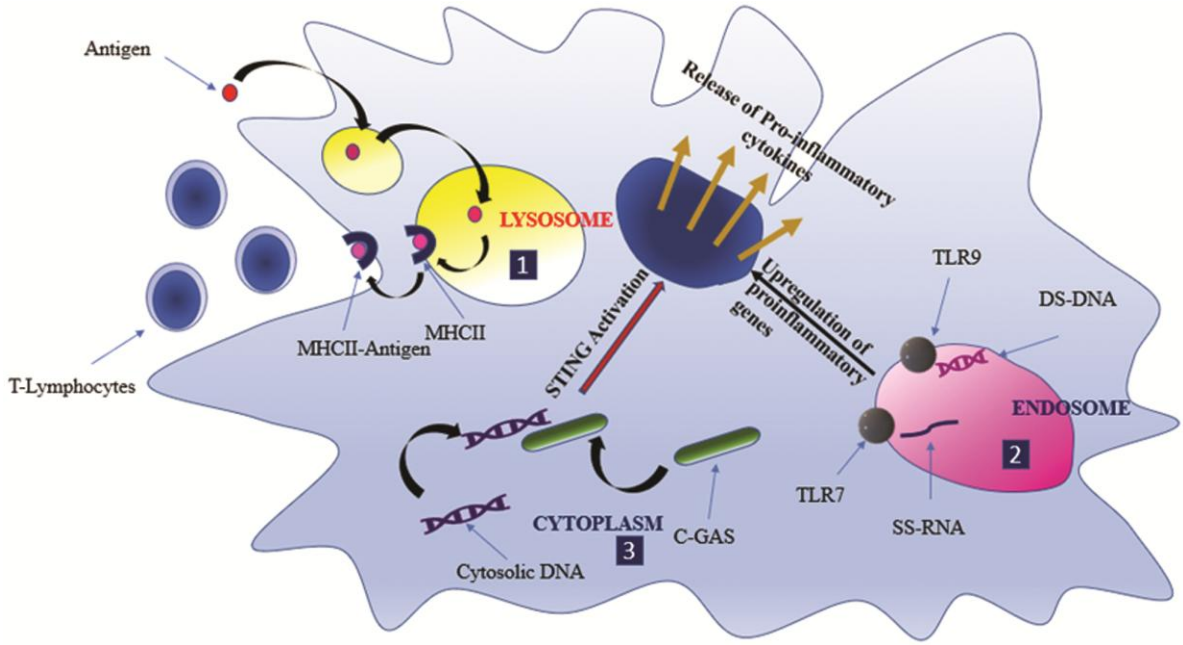

Fig. 3 — Immunomodulatory action of HCQ in antigen presenting cells in SARS-CoV-2 viral infection

The ECG was within the normal limits on both occasions. Other adverse effects were pain abdomen (single episode) and dizziness (disappeared within 12 h).

Few studies on the adverse effects of HCQ in COVID-19 are available in literature. A countrywide web-based study found adverse events in 37. 9% of subjects in a sample size of 166[15]. The most common adverse event was observed from the gastrointestinal system (30. 7%)[15]. In comparison, we reported an adverse event in 14. 55% of the subjects with gastrointestinal adverse effect being observed in 50% (4 out of 8) of all events. Although QT prolongation and ventricular arrhythmia was noticed in few published case reports, no such adverse effects was seen in our study[14,16]. Interestingly, none of the participants in this study showed any symptoms of warranting testing for COVID-19 till 31st July 2020.

Limitations of the study

The limitations of this study includes short duration of the study, the small sample size in a single centre, no follow up with the subjects and without any follow up investigations.

Conclusion

Our study indicates that the prophylactic weekly single dose of hydroxychloroquine is not associated with any serious adverse effects within 1-7 weeks of initiation of therapy. Elucidation of the long term, chronic adverse effects, and immunomodulatory actions, if any, requires further studies.

Acknowledgement

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Conflict of interest

All authors declare no conflict of interest.

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