FIGURES

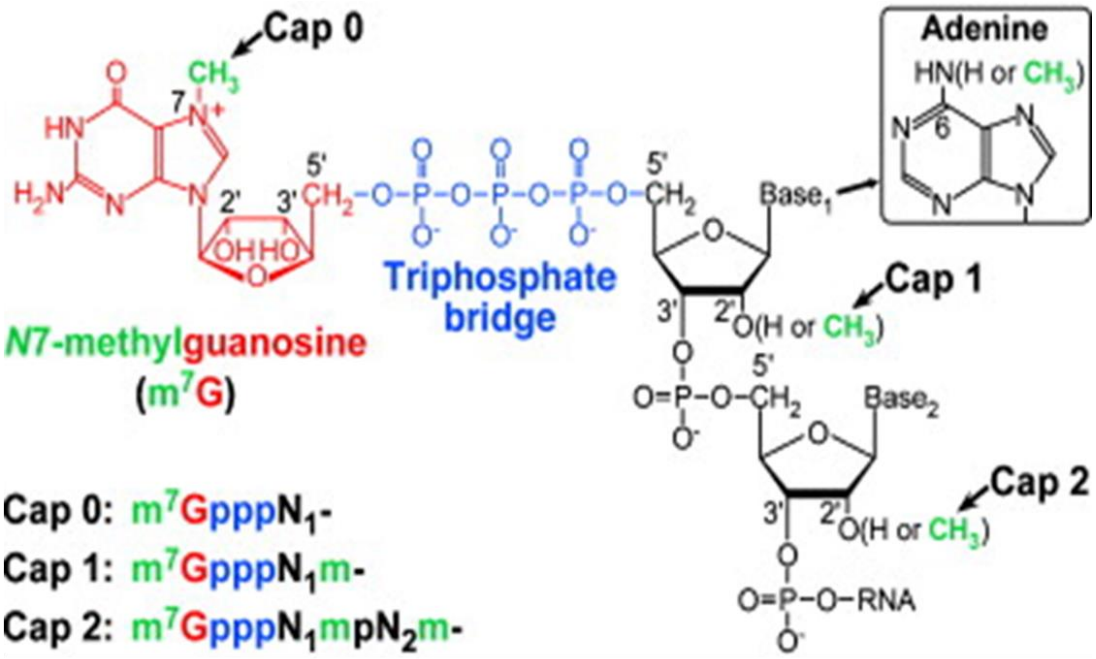

Figure S1: RNA methylation capping

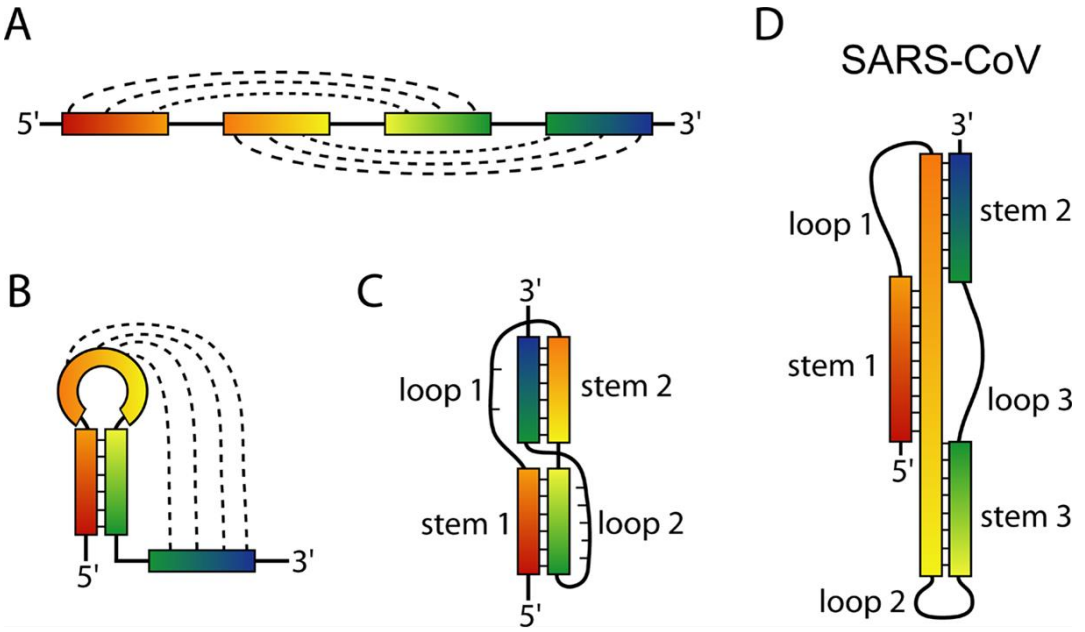

Figure S2: Various types of Pseudoknots of Virus mRNA

https://www.facebook.com/WHO/posts/3061342457244450

Date: 13 April 2020

TO THE HONOURABLE DIGNITARIES

Sir/ Madam:

Subject: **Use of Hydroxychloroquine for treatment of COVID-19**

By Samir Kumar Patra, Department of Life Science, NIT-Rourkela, India.

Summary:

Use of nonspecific drugs would keep the pathogen in dormant stage. Metabolic excretion of the drug after dosage withdrawal may ignite the growth potential of the pathogen and outbreak of the disease.

Explanation:

Since the outbreak of novel coronavirus (2019-nCoV) from Wuhan, China, it spread rapidly, with cases now confirmed initially with mild fever and severe suffocation leading to death in multiple countries. Close contact of the victims with others and coordination between clinicians and public health authorities at the local, state, national and international levels, as well as the need for rapid dissemination of clinical information related to the care of patients with this emerging infection highlighted the importance of additional measures of protection (1, 2).

Considering the efficacy of Chloroquine (CQ) and a less toxic derivative of CQ, Hydroxychloroquine (HCQ) against malaria, some groups around the globe tested its potential against COVID-19 (3-5). SARS-CoV-2 binds sialic acids and gangliosides of host cell membrane with high affinity through its spike (S) protein. It has been demonstrated that the viral spike (S) is no longer able to bind gangliosides in presence of CQ/HCQ (6). These are in vitro studies; while the efforts are praise worthy, we need caution to apply in the clinics.

The basic mode of action of Plasmodium falciparum in human body is; after the carrier mosquito (female Anopheles) bite, the parasite is transported to liver through circulation. In the liver they multiply and released back to circulation it enters into the RBC. In the RBC as well the parasite can grow very well; which results rapid loss of effete RBCs and decrease of other blood cells (see Figure 1), including platelet, lymphocytes and other haematological parameters (7).

As summarised by buffet et al (2011) and Maier et al. (2009), “Infection by early parasite stages (rings), late parasite stages trophozoites and schizonts (mature forms) have substantial differences of adhesion/deformability that explains passage through small vessels. Early stage (rings) parasites have less deformability preventing their circulation in the peripheral blood. Sequestration of mature forms is induced by their adherence to endothelial cells, blood cells, platelets, and uninfected RBCs (uRBCs). These interactions are mediated by multiple

host receptors recognized by parasite adhesins. The major parasite adhesion is the variant P. falciparum Erythrocyte Membrane Protein 1 (PfEMP1) encoded by the var multigene family. The expression of PfEMP1 on mature forms is concentrated at electron-dense knobs on the Infected RBC membrane. At the end of the cycle, schizont rupture releases antigens and components that elicit a strong systemic inflammatory response, which indirectly contributes to pathogenesis by stimulating the expression of inducible endothelial receptors, thereby promoting further cytoadherence. When sequestered, mature forms do not circulate and escape retention and destruction by the spleen. Indeed, mature forms are undeformable and unable to cross interendothelial slits of the spleen. Interestingly, mature forms of P. vivax are more deformable than normal RBCs and are observed in the peripheral circulation. Sequestration of mature forms of P falciparum (i.e., escape from the spleen) indirectly favors parasite multiplication and explains the initial Log-linear increase of parasite loads in nonimmune subjects with P falciparum malaria. In patients with cerebral malaria sequestration of mature forms in small vessels, and total body parasite biomass are greater than in patients with uncomplicated attacks, or other severe attacks" (8,9).

Now the question is how CQ or HCQ may recover the malaria patient? Among the ring- and schizont-stage of P. falciparum metabolic fluxes that govern reduced thioredoxin and phosphoenolpyruvate synthesis are significantly decreased in the ring stage and crucial to chloroquine-based inhibition of P. falciparum DNA replication (10-13).

In vitro demonstration that CQ has multiple mechanisms of action; significantly differs according to the pathogen. CQ/HCQ may inhibit UDP-N-acetylglucosamine 2-epimerases require for biosynthesis of sialic acids. The sialic acids are critical components of ligand (Virus) recognition, by the glycosylated transmembrane proteins of the host cells. The possible interference of CQ with sialic acid biosynthesis could account for the broad antiviral spectrum of that drug since viruses such as the human coronavirus HCoV-O43 and the orthomyxoviruses use sialic acid moieties as receptors. Inhibition of glycosylation of a virus cell surface receptor, by inhibiting angiotensin-converting enzyme 2 (ACE2) is possible by HCQ/CQ (5,6). In the other way, CQ/HCQ can inhibit endosome-mediated viral entry, if Novel corona virus (causing COVID-19) enters the host cells through the route of SARS-CoV-1 after binding of the DC-SIGN receptor. Inhibition of hepatitis A virus by CQ may possibly work by preventing the uncoating of the endosome, thus blocking its entire replication cycle. CQ can inhibit post-translational modifications proteases and glycosyltransferases, essential for viral maturity. For HIV, inhibition of glycosylation of the gp120 envelope glycoprotein causes virus particles non-infectious. In case of influenza virus CQ improve the cross-presentation of non-replicating virus antigen by dendritic cells to CD8+ T-cells. CQ may act on the immune system through cell signalling and regulation of pro-inflammatory cytokines; for example it inhibits phosphorylation (activation) of the p38 mitogen-activated protein kinase (MAPK) in THP-1 cells. Cells MAPK signalling cascade is essential for viruses to achieve their replication cycle (3-6).

Nonspecific drugs against dreadful viruses is always threatening; because, the viruses if not be killed would stay in dormant stages elsewhere in the lymph nodes. After the withdrawal of

the dosages and complete excretion of the drug, CQ/HCQ from the physiological system, including blood through cellular metabolism, the disease would relapse. The dormant viruses within our system would cause outbreak of the disease in near future, and unfortunately enough the situation would be worst because HCQ will not be effective against those drugs.

In view of this and considering the welfare of the society, randomized controlled trials are highly recommended and essential steps for future strategies (14).

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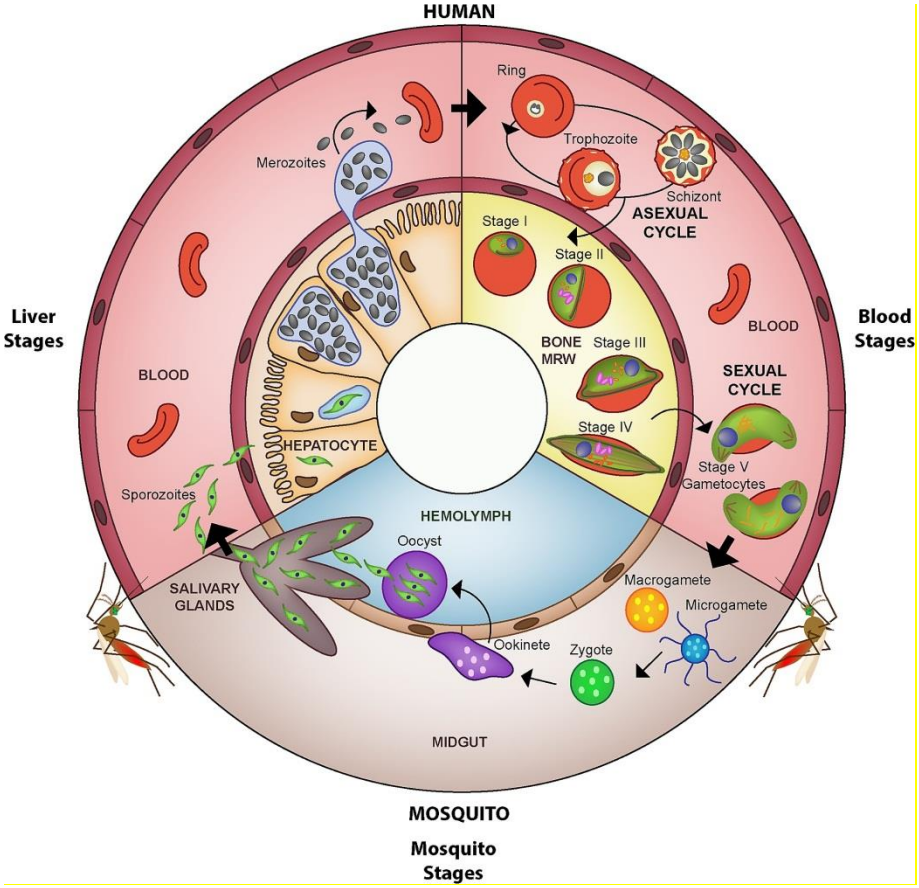
FIGURES

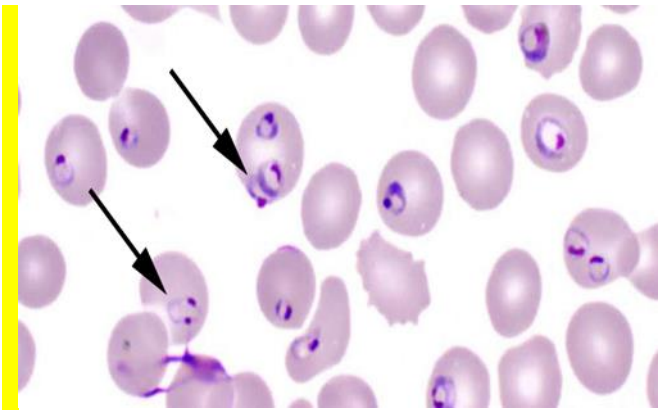
Figure 1. A



Life cycle of Plasmodium falciparum: Contributed by Sandra K. Nilsson, Lauren M. Childs, Caroline Buckee, and Matthias Marti.

Source: https://figshare.com/articles/_Life_cycle_of_Plasmodium_falciparum_/1453753/1

Figure 1. B



Ring form of Plasmodium falciparum: Contributed by Bobbi Pritt, M.D.

Source: http://www.pathologyoutlines.com/topic/parasitologymalariapfalciparum.html

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