



## Can hydroxychloroquine be protective against COVID-19-associated thrombotic events ?

Christian Devaux, Laurence Camoin-Jau, Jean-Louis Mege, Didier Raoult

### ► To cite this version:

Christian Devaux, Laurence Camoin-Jau, Jean-Louis Mege, Didier Raoult. Can hydroxychloroquine be protective against COVID-19-associated thrombotic events ?. Journal of Microbiology, Immunology and Infection, 2021, 54 (1), pp.37-45. 10.1016/j.jmii.2020.12.010 . hal-03281965

**HAL Id: hal-03281965**

**<https://amu.hal.science/hal-03281965>**

Submitted on 10 Mar 2023

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

**Title: Can hydroxychloroquine be protective against COVID-19-associated thrombotic events ?**

Christian A. Devaux<sup>a,b,\*,\*\*</sup>, Laurence Camoin-Jau<sup>a,c,\*</sup>, Jean-Louis Mege<sup>a</sup>, Didier Raoult<sup>a</sup>

<sup>a</sup>Aix-Marseille Univ, IRD, APMH, MEPHI, IHU-Méditerranée Infection, Marseille, France

<sup>b</sup> CNRS, Marseille, France

<sup>c</sup> Laboratoire d'Hématologie, Hôpital de la Timone, APMH, boulevard Jean- Moulin, 13005 Marseille, France

**\*Contributed equally**

**\*\*Corresponding author :**

Christian Devaux, PhD

IHU Méditerranée Infection

19-21 Boulevard Jean Moulin

13385 Marseille, France

Phone: (+33) 4 13 73 20 51

Fax : (+33) 4 13 73 20 52

E-mail: christian.devaux@mediterranee-infection.com

Abstract length: 122 words; Manuscript length: 2852 words ; Figure: 1

**Keywords:** COVID-19; thrombosis; antiphospholipid antibodies; hydroxychloroquine

## Abstract

Although SARS-CoV-2 is considered a lung-tropic virus, severe COVID-19 is not just a viral pulmonary infection, clinically it is a multi-organ pathology with major coagulation abnormalities and thromboembolism events. Recently, antiphospholipid (aPL) antibodies were found increased in a large number of COVID-19 patients. Elevated aPL have been well documented in antiphospholipid syndrome (APS), a systemic autoimmune disorder characterized by recurrent venous or arterial thrombosis and/or obstetrical morbidity. Among treatment regimen of APS, hydroxychloroquine (HCQ) is one of the molecules proposed in the primary prevention of thrombosis and obstetrical morbidity in those patients. Due to its antithrombotic properties documented in APS therapy, HCQ could be considered a good candidate for the prevention of thrombotic events in COVID-19 patients in association with anticoagulant and its repurposing deserves further evaluation.

## Introduction

The 4-aminoquinoline drug hydroxychloroquine (HCQ) belong to the same molecular family than chloroquine (CQ), an amine acidotropic form of natural quinine that was synthesised in the early 1930s and emerged approximately 70 years ago as a substitute for quinine against malaria. HCQ was reported to be as active as CQ against *Plasmodium falciparum* and less toxic, allowing long periods of high dose therapy with very good tolerance<sup>1</sup>. Moreover, HCQ has been widely used for many years in the therapy of autoimmune diseases as it acts as immune modulator interfering with lysosomal activity and autophagy, modulating signaling pathways (such as inhibition of Toll-like receptor signaling in dendritic cells, phospholipase A2 activity and arachidonic acid production in platelets, nitric oxide production by endothelial cells, antiphospholipid- $\beta$ 2 glycoprotein complexes on monocytes surfaces, inhibiting cytokines production by T lymphocytes) and transcriptional activity<sup>2-4</sup>. The antithrombotic properties of HCQ was described as far as 1975<sup>5</sup>. In the context of autoimmune disease (SLE), HCQ inhibits stimulated platelets at the arachidonic acid pathway and thromboxane 2 generation in activated platelets (an activator of platelets aggregation), which is associated with decreased circulating levels of endothelin-1, and allows improvement of endothelial function<sup>3, 6-8</sup>. Interestingly, using network-based approach to prediction and population-based validation of *in silico* drug repurposing it was found that the Healthcare registry data for 220 million people showed that HCQ intake (a series of 37,795 patients receiving HCQ) was associated with a lower risk for coronary artery disease<sup>9</sup>.

The main purpose of this review is to summarize the evidence supporting a potential beneficial role of HCQ in the prevention of thrombosis in patients with antiphospholipid syndrome and discuss the possible repurposing of this molecule as an additional member of the therapeutic arsenal in association with classical antithrombotic drugs used for the prevention of the obstructive thrombo-inflammatory syndrome associated with severe forms of COVID-19.

## The pathophysiology of COVID-19

An outbreak caused by a novel human coronavirus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was first described in Wuhan in December 2019<sup>10-12</sup>. During the past nine months the SARS-CoV-2 has spread worldwide and was responsible for severe COVID-19 disease forms characterized by cytokine storm, acute respiratory distress

syndrome (ARDS), and severe thrombotic events leading to multiple organ dysfunction syndrome (MODS), and high risk of fatal evolution <sup>11,13-14</sup>. To date (one year after the first outbreak in China), SARS-CoV-2 has been responsible for more than 1.57 million deaths among about 68.95 million of infected people (<https://coronavirus.jhu.edu/map.html>), and these numbers are still raising daily.

It is currently admitted that COVID-19 pathogenesis is mainly characterized by production of pro-inflammatory cytokines including IL-6, a key contributor in the development of cytokine storm associated with microvascular injury, obstructive thrombo-inflammatory syndrome which represent the primary causes of lethality <sup>15,16</sup>, with an estimated fatality rate of 2,27%. The lack of specific treatment for COVID-19 led all clinical teams to try to speed up the implementation of therapeutic strategies by carrying out drug repurposing. HCQ which was known to inhibit the replication of several coronavirus *in vitro*, was reported to inhibit SARS-CoV-2 *in vitro* <sup>17-20</sup>. This antiviral activity likely occurs through several mechanisms and cellular targets <sup>21-22</sup>. By February 2020, the first report from China on the clinical efficacy of chloroquine phosphate in treatment of COVID-19 associated pneumonia was submitted for publication <sup>23</sup>. Considering the possible benefit of this molecule as both antiviral agent and immunomodulator compound, it was tempting to evaluate its potential in the prevention and/or treatment of COVID-19 <sup>24-28</sup>. This therapeutic approach has lead to an impassioned global debate on the promises and pitfalls of HCQ in COVID-19, in both clinical and scientific communities. By mid-July 2020, among 2,654 registered clinical trials from 43 countries aimed at testing the capacity of compound at preventing severe COVID-19, 239 included HCQ treatment or prophylaxis while 82 addressed chloroquine efficiency (Clinicaltrials.gov). Still today, the debate remains fierce between those who consider HCQ to save lives, those who find no significant impact on COVID-19 progression and those who still claim that HCQ is toxic <sup>29-34</sup>. This does not make it possible to conclude whether HCQ is beneficial or not, it simply indicates that the case series and treatment protocols are different and that meta-analysis algorithms are either unsuitable or misused.

As knowledge about the disease grew it became evident that one of the major issues to be addressed in severe COVID-19 was that of thrombosis <sup>35,36</sup>. The use of anticoagulants was considered as a major therapy to reduce the harmful circle of inflammation-coagulation observed in patients with a severe form of COVID-19 <sup>37,38</sup>. Antithrombotic therapy improved COVID-19 patients outcomes <sup>39</sup>.

*In vitro*, HCQ was found to induce attenuation of human aortic endothelial cells activation upon exposure to proinflammatory cytokine TNF $\alpha$  by reducing of VCAM and IL-1 $\beta$  production <sup>9</sup>. The ectonucleoside triphosphate diphosphohydrolase I/CD39, present on endothelial cells and circulating blood cells such as leukocytes, neutrophils, T- and B-lymphocytes and macrophages is a known interface between vascular inflammation and thrombosis through regulation of ATP, ADP and AMP levels <sup>40</sup>. Mice lacking CD39 expression have marked fibrin deposit in pulmonary and cardiac tissues <sup>41</sup>. IL-6 and other cytokines are increased in COVID-19 patients <sup>38, 42-43</sup>, and the profound inflammatory state of the patients can be characterized by high levels of C reactive protein (CRP) and fibrinogen. These observations led to the adoption of the anti-IL6 receptor monoclonal antibody tocilizumab for the treatment of pneumonia-associated to cytokine storm <sup>44, 45</sup>. It is also known that blocking IL-6 increases the frequency of CD39+ Treg cells <sup>46</sup>. In severe COVID-19, patients with thrombosis have significantly higher blood levels of markers of neutrophil extracellular traps (NETs), neutrophil activation (calprotectin, cell-free DNA) and D-dimers <sup>47</sup>. SARS-CoV-2 induces functional changes in platelet. Platelet hyperreactivity may contribute to thrombotic events through increased platelet-platelet and platelet-leukocytes interactions <sup>48</sup>. Moreover, antiphospholipid (aPL) antibodies were recently found increased in a large number of COVID-19 patients <sup>49-54</sup>. Higher aPL antibodies were associated with neutrophil hyperactivity including the release of NETs <sup>55</sup>. Recently, it was reported that increased count of CD15<sup>+</sup>CD16<sup>+</sup> neutrophils is a COVID-19 signature <sup>56</sup>.

### **Thrombotic disease in severe COVID-19 patients**

Coagulation abnormalities are common in COVID-19 patients. Degree of elevation of fibrinogen and D-Dimers is correlated to the severity of the disease and elevated D-dimer upon admission and during course of disease is associated with increased mortality <sup>57-61</sup>. Clinical evidence indicated that COVID-19 is associated with an increase risk of thrombotic events leading to sustained activation of the clotting, generally venous thromboembolism (VTE) almost assuredly underdiagnosed, due to the difficulty of the performing blood vessels investigation when patients are under isolated care and the fact that the D-dimer level is already high. However, 20-30 % of acute pulmonary embolisms were reported in critically patients <sup>15, 54, 62-65</sup>. Large vessel occlusion stroke was also described <sup>66, 67</sup>. While diagnosis of disseminated intravascular coagulopathy (DIC) has been mainly discussed in severe COVID-19, this

diagnosis is limited to end stage of COVID-19. Yet, this disorder is not a typical DIC  
fibrinogen levels are often high and platelets are rarely reduced. It is more similar to  
complement mediated thrombotic microangiopathy (TMA) syndromes, that is involve  
disorders of complement. Mediators of TMA syndromes overlap with those released in  
cytokine storm <sup>68, 69</sup>. Interestingly, the presence of platelet-fibrin microthrombi in small  
arterial vessels of lung tissues was reported in 87% (33/38) of patient who have died with  
COVID-19 and for whom pulmonary post-mortem examination was requested <sup>70</sup>. For the  
patients who have had symptomatic forms of the disease, delayed pulmonary fibrosis may be  
found in a relatively important proportion of COVID-19 patients once they have healed <sup>37</sup>.  
These observations allowed proposing a new pulmonary vasculopathy named pulmonary  
intravascular coagulopathy (PIC) <sup>43, 71</sup>. Moreover, microangiopathic vessel occlusions and  
endothelium damages was described in kidney <sup>72</sup>. Elevated plasma von Willebrand factor  
(VWF) concentrations in COVID-19 patients, a factor mainly biosynthesized by activated  
endothelial cells, was reported <sup>73-75</sup>. Deposits of complement components C5b-9, C4d in the  
microvasculature of lung and skin was also reported <sup>76</sup>. These observations would be in favor  
of complement activation which participates to microvascular injury.

Also, COVID-19 induced micro and microvasculature dysfunction. The cytokine storm  
observed plays a determining role in this immunoinflammatory thrombosis and in endothelial  
damages. All of these mechanisms, whose kinetics remains unclarified today, might explain  
the occurrence of various thromboembolic events and multiples organ dysfunction observed  
in critical COVID-19 patients.

In parallel to these observations, it is important to underline the fact that a high prevalence of  
antiphospholipid (aPL) antibodies has been observed in critical COVID-19 patients and is  
reminiscent of a clinical scenario of antiphospholipid syndrome (APS). Among aPL  
antibodies, Lupus anticoagulant (LA) are more associated to thrombotic events. In agreement  
with Helms' study <sup>54</sup>, it was demonstrated that 85 % of critically COVID-19 patients  
presented Lupus anticoagulant <sup>77</sup>. Recently, our group described a prevalence of 62% and  
25% of LA respectively in hospitalized and ambulatory patients (Camoin-Jau et al.,  
Submitted). This high prevalence could be linked to cytokine storm and dysimmunity. To the  
best of our knowledge, the role of Lupus anticoagulant in the occurrence of thrombotic event  
is not yet demonstrated. High levels of aPL- $\beta$ 2GPI antibodies where also recently found in a  
large number of COVID-19 patients <sup>52, 78</sup>. Regarding clinical events described during COVID-

19 infection and biological abnormalities, Cavalli and colleagues <sup>79</sup>, suggested that a secondary form of APS is present in COVID-19 patients.

### **Lesson from the Antiphospholipid syndrome (APS)**

Mainly associated with systemic lupus erythematosus (SLE) and other autoimmune diseases (such as rheumatoid arthritis, dermatomyositis, systemic scleritis, Sjögren's syndrome), the antiphospholipid syndrome (APS) is a systemic autoimmune disorder characterized by recurrent venous or arterial thrombosis with or without pregnancy morbidity in the presence of persistent antiphospholipid (aPL) autoantibodies including the lupus anticoagulant (LA) or, anticardiolipin (aCL) or anti-beta 2glycoprotein (anti $\beta$ 2-GPI) autoantibodies. In 53% of patients it exists alone as primary APS (PAPS) <sup>80, 81</sup>. The presence of antibodies directed against CL and/or  $\beta$ 2-GPI has been observed in a percentage of healthy individuals without clinical symptoms of APS ranging from 4.5 to 5.5% <sup>82, 83</sup>. There is still controversy about whether pharmacologic primary thromboprophylaxis is indicated in asymptomatic carriers of aPL antibodies <sup>84</sup>.

Many clinical trials have been focused on the benefit of aspirin on prevention of thrombotic events in APS patients. Actually, thromboprophylaxis with aspirin is proposed only for asymptomatic aPL carriers with high risk profile, especially in the presence of other thrombotic risk factors <sup>85, 86</sup>. Low molecular weight heparin are proposed in all aPL carriers in high-risk situations, such as surgery, prolonged immobilization and the puerperium. Usually APS patient receive oral anticoagulant <sup>87-89</sup>, and/or low-dose aspirin <sup>90</sup>.

By opposite, HCQ (grade 1B recommendation) and low-dose aspirin are proposed in SLE with LA or isolated persistent aCL <sup>86</sup>. Indeed, several studies suggest that beside its anti-inflammatory effects, HCQ could play a role in lowering antiphospholipid titers and potentially be anti-thrombotic through several mechanisms involving inhibition of platelet adhesion, intravascular aggregation of red blood cells, interactions between platelets and coagulation factors, and binding of antiphospholipid (aPL) antibodies phospholipid surface <sup>91-93</sup>. In antiphospholipid syndrome (APS) an important risk of thrombosis relapse should be considered <sup>94</sup>. HCQ benefit to protect against thromboembolism was described more than 30-years ago, with reported reduction of erythrocyte aggregation *in vitro* and thrombus size <sup>95, 96</sup>. *In vitro*, CQ at 1 mM inhibit both ADP-stimulated platelets aggregation and thrombin-stimulated platelets aggregation <sup>97</sup>. Jancinova and colleagues hypothesized that CQ, as a



cationic amphiphilic molecule, might inhibit platelets aggregation either through interaction with membrane phospholipids, membrane receptor such as ADP receptor, induce the displacement of membrane-bound calcium, or pass membrane and directly act on platelets phospholipase A2, phospholipase C or calmodulin, or the production of aggregating-amplifying substances such as histamine, serotonin and adenine nucleotides. HCQ administration to patients was found to significantly reduce the thrombus size and duration <sup>98</sup>. Similar observations were reported using mouse models of APS in which administration of HCQ limited aPL binding on target cells decreased pro-inflammation, and reduced the size and duration of the thrombus <sup>98, 99</sup>. Miranda and colleagues <sup>99</sup>, found that HCQ increased the p-eNOS/e-NOS ratio leading to an improvement in the production of NO. A protective effect of HCQ against thrombosis in asymptomatic aPL-positive APS individuals was reported <sup>100</sup>. In the LUMINA observational cohort of 442 SLE patients HCQ was found thromboprotective <sup>101</sup>. These encouraging preliminary reports lead to the organization of an international consortium (APS ACTION) with the aim to set up a randomized controlled trial of HCQ in the primary thrombosis prevention of persistently aPL-positive but thrombosis free patients without other systemic autoimmune diseases <sup>102</sup>; they had the objective to investigate a cohort of 1,000 patients age 18-60 years, without pharmaceutical support, but 6-years only 20 patients had been included in the cohort and the trial was stopped <sup>103</sup>. However, there are evidences that HCQ could be beneficial to APS patients. Among asymptomatic aPL-positive patients with SLE, primary prophylaxis with HCQ appears to reduce the frequency of thrombotic events <sup>104</sup>. HCQ could reduce subclinical atherosclerosis and its use may provide survival benefit <sup>105</sup>. HCQ was found to protect patients against both venous and arterial thromboses <sup>106</sup>. HCQ is considered to have several antithrombotic effects <sup>107-110</sup>. Rand and colleagues <sup>108, 109</sup>, demonstrated that HCQ reduces the binding of aPL autoantibodies/ $\beta$ 2GPI complexes (responsible for the thrombotic effect) to phospholipid bilayers, and protect the anticoagulant annexinA5 (AnxA5) from disruption by aPL autoantibodies. Interestingly, HCQ blocks platelet aggregation and adhesion and improves cholesterol profiles <sup>111</sup>. Schmidt-Tanguy and colleagues <sup>112</sup>, reported that in patients with clinical history of venous thrombosis (one or two episodes) 6/20 patients treated with oral anticoagulants (fludione) alone showed recurrent venous thromboses whereas no recurrent venous thromboses (0/20) in patients who received HCQ (400 mg daily) in addition to oral anticoagulants (fludione). The long term administration of HCQ to PAPS patients reduced aPL antibodies <sup>92</sup>. HCQ also partially reverse the aPL-induced impaired trophoblast migration <sup>113</sup>. This is consistent with the observation that addition of HCQ to conventional treatment may be associated with a

reduction of first-trimester miscarriages in pregnant patients <sup>114</sup>.

Trials are still ongoing. The HYPATIA multicenter trial will examine the use of HCQ versus placebo in aPL-positive women planning to conceive <sup>115</sup>. A more recent study, HIBISCUS, plans to examine the effect of HCQ on secondary thrombosis and APS-related morbidity in PAPS <sup>116</sup>. HCQ was also reported to decrease LDL cholesterol and serum glucose levels in PAPS patients likely contributing to reduce the risk of thrombosis <sup>117</sup>. Recently, a pilot open label randomized prospective study on the use of HCQ (200 mg daily for patients weighing below 60 Kg and 400 mg daily for those weighing above 60 Kg) for prevention of thrombosis in 50 patients with PAPS concluded to the efficiency of the treatment with a decreased incidence of thrombosis associated to a reduction in aPL titers <sup>118</sup>.

## **Anti-thrombotic effect of HCQ in COVID-19 ?**

### **HCQ as a possible treatment of thrombotic events observed in COVID-19?**

Right at the center of the debate on the use of HCQ as a prophylaxis and treatment of COVID-19 and lack of proper design of many trials, the idea that treatment of COVID-19 patients by HCQ might reduce the thrombotic events observed in severe forms of the disease has emerged <sup>119</sup>.

The vascular endothelium functions as an integral barrier, separating blood from the subendothelial tissue compartments. It maintains its integrity and blood fluidity by acting as an anticoagulant through suppression of platelets activation and induction of fibrinolysis. Anti-SARS-CoV-2 defense mechanisms are likely to activate a tissue aggressive immune response including exaggerated IL-6 production that drives inflammatory reactions and the so called "cytokine storm" that increases tissue damage. Coagulopathy and vasculopathy walls following a pro-inflammatory process result in rapid activation of mechanisms aimed at leading to local damage repair, immune cells accumulation to prevent infection, platelets aggregation for primary and secondary hemostasis.

Thrombosis is common during critical illness especially in the oldest patients who had preexisting cardiovascular disease and it was reported in COVID-19 patients <sup>49, 52, 54</sup>. There is currently a debate whether or not the number of patients who experience arterial thrombotic events in COVID-19 is higher rate compared to critically ill patients without SARS-CoV-2

carriage <sup>120</sup>. Whatever the answer, COVID-19 appears to induce a hypercoagulable state with elevated fibrinogen and the fact remains that clinicians must prevent and/or treat thrombosis. In patients with previous thrombosis attributable to APS, anticoagulation has formed the cornerstone of treatment. Inflammation in the presence of aPL, is associated with an increased thrombotic risk, that requires rapid treatment with anticoagulant to reduce mortality. This is very similar to what is observed in COVID-19 (inflammation and thrombosis), which suggests that in this pathology also taking HCQ could have a beneficial effect against thrombotic events and preliminary investigation of such therapy is under evaluation <sup>121</sup>.

## Conclusion

If the therapeutic use of HCQ in COVID-19 is still the subject of passionate debate in the infectious disease community, rheumatologists have successfully used it for a long time for the prevention of pro-inflammatory process and the thrombosis, two pathological processes frequently encountered in severe cases of COVID-19. When the HCQ/azithromycin was initiated in Marseille IHU Méditerranée infection for the treatment of people who were found positive for SARS-CoV-2, the main objective of this protocol was to take advantage of both the antiviral and immunosuppressive properties of HCQ <sup>122</sup>. Yet, we might hypothesize that the low mortality of COVID-19 patients treated in Marseille IHU with HCQ (fatality rate estimated 0.4% to be compared to 2.27% worldwide) could be also due to a protective effect of HCQ against thrombosis when used in association with a well-chosen anticoagulant. This will require further evaluation.

## **Funding**

This work was supported by the French Government under the « Investissements d’avenir » (Investments for the Future) program managed by the Agence Nationale de la Recherche (French ANR: National Agency for Research), (reference: Méditerranée Infection 10-IAHU-03), the Région Provence Alpes Côte d’Azur and European funding FEDER PRIM

## **Authorship**

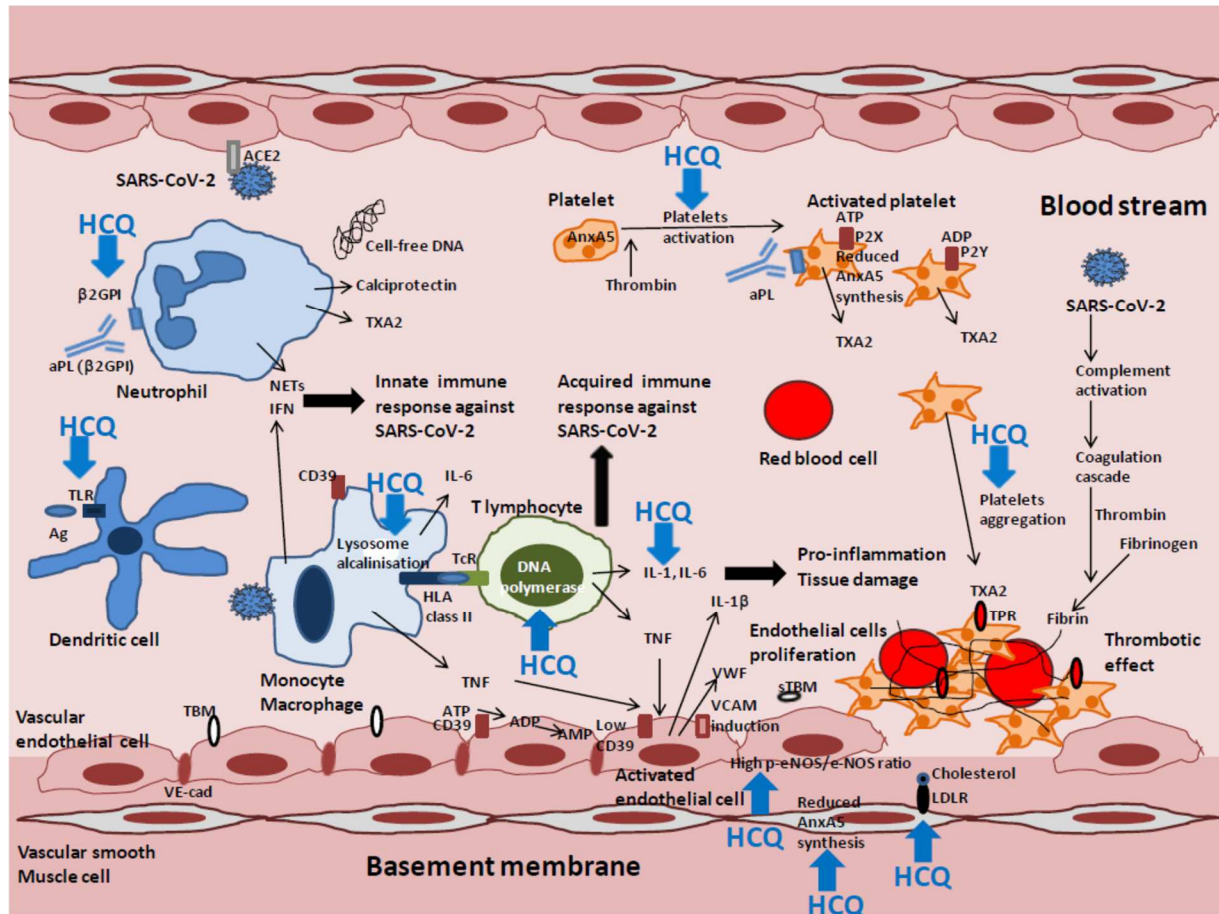
All authors contributed to the design of the study and conceived the manuscript. CD and LCJ wrote the paper. DR obtained the funding for this study. All authors reviewed and approved the final version of the manuscript

## **Declaration of Competing Interest**

CD declares a link of interest with the Sanofi pharmaceutical company which markets hydroxychloroquine. The other authors (LCJ, JLM, and DR) declare that they have no competing interests.

**Figure 1:** Possible mechanisms of action of hydroxychloroquine (HCQ) against COVID-19-associated thrombosis. The vascular endothelium functions as an integral barrier through myriad mechanisms including VE-cadherin, and maintains blood fluidity by acting as an anticoagulant through suppression of platelets activation and induction of fibrinolysis, mechanism including heparan sulfate proteoglycans and CD39. During SARS-CoV-2 infection, innate and acquired immune defense mechanisms are activated (including the cytokine storm IL-1, IL-2, IL-6, IL-8, IL-17, TNF $\alpha$ ) that provokes tissue damage in the lung parenchyma and the immediately adjacent bronchial alveolar lymphoid tissue and disruption of blood vessel walls. The endothelial cells express the angiotensin I converting enzyme 2 (ACE2) molecule that act as cell-surface-receptor that facilitates SARS-CoV-2 entry into these cells. When activated by proinflammatory cytokines, or neutrophil extracellular traps, endothelial cells produce von Willebrand factor that retains platelets and leucocytes to the vessel wall and activates coagulation systems resulting in rapid activation of mechanism aimed at leading to local damage reparation, immune cells accumulation to prevent infection, platelets aggregation for primary and secondary hemostasis. The hyper-reaction set up in response to vascular damage, can influence a propensity toward local vascular micro-thrombosis. COVID-19 patients suffer from prominent alveolar oedema, intra-alveolar proteinosis, cell infiltration including lymphocytes apoptosis of virally-infected pneumocytes, fibrin deposition. HCQ treatment of COVID-19 patient is likely to reduce pro-inflammation and vascular micro-thrombosis due to its multiple actions (the expected mechanisms of action of HCQ to counteract pro-inflammation and thrombosis are indicated by a blue arrow and HCQ) in addition to reduce the patient viral load. aPL: antiphospholipids; IL-6: interleukin-6; TLR: toll like receptor; Ag : antigen; IFN: interferon; TNF: tumor necrosis factor; CD39/ENTPD1: Ectonucleoside triphosphate diphosphohydrolase-1 (also known as or P2 receptors: P2X receptors are ion channels that open upon binding of ATP; P2Y receptors mediate cellular response to purine and pyrimidine such as ATP, ADP, UTP; in physiological conditions CD39 catalyze the reduction of ATP and ADP pool to AMP and CD73 transform AMP to adenosine whereas nucleotides released during cell activation/injury bind to P2 receptors to activate thrombo-inflammatory programs); NETs: neutrophil extracellular traps; TXA2: Thromboxane A2 (induce platelets aggregation); AnxA5: annexinA5 (or annexin V or anchorin CII; anticoagulant, interact with phospholipids); TPR : thromboxane A2 prostanoid receptor: VE-cad: VE-cadherin; TBM: thrombomodulin prevents thrombosis; upon endothelial cell activation a soluble form of TBM (sTBM) is released in plasma further promoting procoagulant mechanisms. VWF: von Willebrand factor; Fibrin: fibrin is formed

from blood plasma fibrinogen (produced in the liver) by the action of thrombin; red thrombus is composed of erythrocytes enmeshed in a fibrin network; LDLR: Low density lipoprotein receptor (bind LDL/cholesterol).



## References

1. Wellems TE, Plowe CV. Chloroquine-resistant malaria . *J Infect Dis*, 2001; **184**: 770-776.
2. Xourgia E, Tektonidou MG. Type I interferon gene expression in antiphospholipid syndrome : pathogenetic, clinical and therapeutic implications. *J Autoimmun* 2019; **104**: 102311
3. Schrezenmeier E, Dorner T. Mechanisms of action of hydroxychloroquine and chloroquine: implications for rheumatology. *Nat Rev Rheumatol* 2020; **16**: 155-166
4. Vitte J, Moïse M, Mezouar S, Bailo Diallo A, Boumaza A, Mege JL, Desnues B. Immune modulation as a therapeutic option during the SARS-CoV-2 outbreak: the case for antimalarial aminoquinolones. *Front Immunol* 2020a; **11**: 2159
5. Genton E, Gent M, Hirsh J, Harker LA. Platelet-inhibiting drug in the prevention of clinical thrombotic disease (third of three parts). *N Engl J Med* 1975; **293**: 1296-1300
6. Nosal R, Jancinova V, Petrikova M. Chloroquine inhibits stimulates platelets at the arachidonic acid pathway. *Thromb Res* 1995; **77**: 531-542
7. Rahman R, Murthi P, Singh H, Gurusinge S, Mockler JC, Lim R, et al. The effects of hydroxychloroquine on endothelial dysfunction. *Pregnancy Hypertens* 2016; **6**: 259-262
8. Li R, Lin H, Ye Y, Xiao Y, Xu S, Wang J, et al. Attenuation of antimalarial agent hydroxychloroquine on TNF-alpha-induced endothelial inflammation. *Int Immunopharmacol* 2018; **63**: 261-269.
9. Cheng F, Desai RJ, Handy DE, Wang R, Schneeweiss S, Barabasi AL, et al. Network-based approach to prediction and population-based validation of in silico drug repurposing. *Nat Commun* 2018; **9**: 2691
10. Lu R, Zhao X, Li J, Niu P, Yang B, Wu W, et al. Genomic characterization and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. *Lancet* , 2020; **395**: 565-574
11. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. *N Engl J Med* 2020; **382**:727–733
12. Frutos R, Lopez Roig M, Serra-Cobo J, Devaux CA. COVID-19: The conjunction of events leading to the coronavirus pandemic and lessons to learn for futur threats. *Front Med* 2020; **7**: 223
13. Huang C, Wang Y, Li X, Ren PL, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* 2020; **395**(10223): 497-506
14. Qin C, Zhou L, Hu Z, Zhang S, Yang S, Tao Y et al. Dysregulation of immune response in patients with COVID-19 in Wuhan, China. *Clin Infect Dis* 2020; **71**(15): 762-768

- 416 15. Middeldorp S, Coppens M, van Haaps TF, Foppen M, Vlaar AP, Miller MCA et al.  
417 Incidence of venous thromboembolism in hospitalized patients with COVID-19. *J Thromb*  
418 *Haemost* 2020; **18**(8): 1995-2002
- 419 16. AbdelMassih AF, Kamel A, Mishriky F, Ismail HA, El Qadi L, Malak L, et al. It is  
420 infection or rather vascular inflammation? Game-changer insights and recommendations from  
421 patterns of multi-organ involvement and affected subgroups in COVID-19. *Cardiovasc.*  
422 *Endocrinol. Metab.* 2020; **9**(3): 110-120.
- 423 17. Savarino A, Boelaert JR, Cassone A, Majori G, Cauda R. Effects of chloroquine on viral  
424 infections: an old drug against today's diseases? *Lancet Inf Dis* 2003; **3**: 722-727
- 425 18. Kearney JE. Chloroquine as a potential treatment and and prevention measure for the  
426 2019 novel Coronavirus: A review. Preprint 17 march 2020, not peer-reviewed. doi:  
427 10.20944/preprints202003.0275.v1
- 428 19. Wang M, Cao R, Zhang L, Yang X, Liu J, Xu M. et al. Remdesivir and chloroquine  
429 effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Res*  
430 2020; **30**: 269-271.
- 431 20. Liu J, Cao R, Xu M, Wang X, Zhang H, Hu H et al. Hydroxychloroquine, a less toxic  
432 derivative of chloroquine, is effective in inhibiting SARS-CoV-2 infection in vitro. *Cell*  
433 *Discovery* 2020; **6**: 16
- 434 21. Hu TY, Frieman M, Wolfram J. Insights from nanomedicine in chloroquine efficacy  
435 against COVID-19. *Nature Nanotech* 2020; [epub ahead of print]. doi:10.1038/s41565-020-  
436 0674-9
- 437 22. Devaux CA, Rolain JM, Colson P, Raoult D. New insights on the antiviral effects of  
438 chloroquine against coronavirus: what to expect for COVID-19, *Int J Antimicrob Agents*  
439 2020a; **55**: 105938
- 440 23. Gao J, Tian Z, Yang X. Breakthrough: chloroquine phosphate has shown apparent  
441 efficacy in treatment of COVID-19 associated pneumlonia in clinicla studies. *Biosci Trends*  
442 2020; [epub ahead of print] doi: 10.1038/s41422-020-0282-0
- 443 24. Li L, Li R, Wu Z, Yang X, Zhao M, Liu J, Chen D. Therapeutic strategies for critically ill  
444 patients with COVID-19. *Ann Intens Care* 2020; **10**: 45
- 445 25. Gautret P, Lagier JC, Parola P, Hoang VT, Meddeb L, Mailhe M., et al.  
446 Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label  
447 non-randomized clinical trial. *Int J Antimicrob Agents* 2020; **56**(1): 105949
- 448 26. Gautret P, Lagier JC, Parola P, Hoang VT, Meddeb L, Sevestre J, et al. Clinical and  
449 microbiological effect of a combination of hydroxychloroquine and azithromycin in 80  
450 COVID-19 patients with at least a six-day follow-up: a pilot observational study. *Travel Med*  
451 *Infect Dis* 2020; **49**: 215219.



452 27. Million M, Lagier JC, Gautret P, Colson P, Fournier PE, Amrane S. et al. Early treatment  
453 of COVID-19 patients with hydroxychloroquine and azithromycin: a retrospective analysis of  
454 1061 cases in Marseille, France. *Travel Med Inf Dis* 2020; **35**: 101738

455 28. Lagier JC, Million M, Gautret P, Colson P, Cortaredona S, Giraud-Gatineau A, et al.  
456 Outcomes of 3,737 COVID-19 patients treated with hydroxychloroquine/azithromycin and  
457 other regimens in Marseille, France. A retrospective analysis. *Travel Med Infect Dis* 2020; **36**:  
458 101791

459 29. Million M, Gautret P, Colson P, Roussel Y, Dubourg G, Fenollar F. Breaking news: the  
460 efficacy of Chloroquine derivatives in COVID-19: a meta-analysis on the first available  
461 reports. *IHU Méditerranée infection* 2020. Preprint not peer-reviewed, IHU prepublication  
462 site on COVID-19

463 30. Sarma P, Kaur H, Kumar H, Mahendru D, Avli P, Bhattacharyya A, et al. Virological and  
464 clinical cure in COVID-19 patients treated with hydroxychloroquine. A systematic review and  
465 meta-analysis. *J Med Virol* (2020) [Epub ahead of print] doi:10.1002:jmv.25898

466 31. Singh AK, Singh A, Singh R, Misra A. Hydroxychloroquine in patients with COVID-19.  
467 A systematic review and meta-analysis. *Diabetes Metab Syndr* 2020; **14**(4): 589-596

468 32. Fiolet T, Guihur A, Rebeaud M, Mulot M, Peiffer-Smadja N, Mahamat-Saleh Y. Effect of  
469 hydroxychloroquine with or without azithromycin on the mortality of COVID-19 patients: a  
470 systematic review and meta-analysis. *Clin Microbiol Inf* 2020. [epub ahead of print] doi:  
471 10.1016/j.cmi.2020.08.022.

472 33. Maisonnasse P, Guedj J, Contreras V, Behillil S, Solas C, Marlin R, et al.  
473 Hydroxychloroquine use against SARS-CoV-2 infection in non-human primates. *Nature*  
474 2020; [epub ahead of print]. doi: 10.1038/s41586-020-2558-4

475 34. The RECOVERY Collaborative Group. Effect of hydroxychloroquine in hospitalized  
476 patients with COVID-19. *N Engl J Med* 2020; **383**: 2030-2040

477 35. Devaux CA, Rolain JM, Raoult D. ACE2 receptor polymorphism : susceptibility to  
478 SARS-CoV-2, hypertension, multi-organ failure, and COVID-19 disease outcome. *J*  
479 *Microbiol Immunol Infect* 2020; **53**(3): 425-435

480 36. Fang L, Karakiulakis G, Roth M. Are patients with hypertension and diabetes mellitus at  
481 increased risk for COVID-19 infection? *Lancet Resp Med* 2020; **8**: e21

482 37. Wang J, Wang BJ, Yang JC, Wang MY, Chen C, Luo GX et al. Research advances in the  
483 mechanism of pulmonary fibrosis induced by Coronavirus disease 2019 and the  
484 corresponding therapeutic measures [Article in Chinese]. *Zhonghua Shao Shang Za Zhi* 2020;  
485 **36**(8):691-697.

486 38. Tang Y, Liu J, Zhang D, Xu Z, Ji J, Wen C. Cytokine storm in COVID-19: the current  
487 evidence and treatment strategies. *Front Immunol.* (2020) [Epub ahead of print]  
488 doi:10.3389/fimmu.2020.01708

489 39. Zhai ZC, Chen Y, Gerotziafas G, Zhang Z, Wan J, et al. Prevention and Treatment of  
490 Venous Thromboembolism Associated with Coronavirus Disease 2019 Infection: A  
491 Consensus Statement before Guidelines. *Thromb Haemost* 2020; **120**(6): 937–948

492 40. Kanthi YM, Sutton NR, Pinsky DJ. CD39: interface between vascular thrombosis and  
493 inflammation. *Curr Atheroscler Rep* 2014; **16**(7): 425

494 41. Enjyoji K, Sevigny J, Lin Y, Frenette PS, Christie PD, Esch JS, et al. Targeted disruption  
495 of cd39/ATP diphosphohydrolase results in disordered hemostasis and thromboregulation.  
496 *Nat Med* 1999; **5**(9):1010-1017

497 42. Grifoni E, Valoriani A, Cei F, Lamanna R, Gelli AMG, Ciambotti B, et al. Interleukin-6  
498 as prognosticator in patients with COVID-19. *J Infect* 2020; **81**(3): 452-482.

499 43. McGonagle D, Sharif K, O'Regan A, Bridgewood C. The role of cytokines including  
500 interleukin-6 in COVID-19 induced pneumonia and macrophage activation syndrome like  
501 disease. *Autoimmunity Rev* 2020; [epub ahead of print] doi:10.1016/j.autrev.2020.102537

502 44. Xu X, Han M, Li T, Sun W, Wang D, Fu B et al. Effective treatment of severe COVID-19  
503 patients with tocilizumab. *Proc Natl Acad Sci USA* 2020; **117**(20) :10970-10975

504 45. Zhang C, Wu Z, Li JW, Zhao H, Wang GQ. Cytokine release syndrome in severe  
505 COVID-19: interleukin-6 receptor antagonist tocilizumab may be the key to reduce mortality.  
506 *Int J Antimicrob Agents* 2020; **55**(2): 105954

507 46. Thiolat A, Semerano L, Pers YM, Biton J, Lemeiter D, Portales P, et al. Interleukin-6  
508 receptor blockade enhances CD39+ regulatory T cell development in rheumatoid arthritis  
509 and in experimental arthritis. *Arthritis Rheumatol* 2014; **66**(2): 273-283

510 47. Zuo Y, Zuo M, Yalavarthi S, Gockman K, Madison JA, Shi H, Woodard W, Lezak SP,  
511 Lugogo NL, Knight JS, Kanthi Y. Neutrophil extracellular traps and thrombosis in COVID-  
512 19. *MedRxiv* (2020) , preprint not peer-reviewed. doi: 10.1101/2020.04.30.20086736

513 48. Manne BK, Denorme F, Middleton EA, Portier I, Rowley JW, Stubben CJ, et al. Platelet  
514 gene expression and function in COVID-19 Patients. *Blood* 2020; [epub ahead of print]. doi:  
515 10.1182/blood.2020007214

516 49. Harzallah I, Debliquis A, Drenou B. Lupus anticoagulant is frequent in patients with  
517 Covid-19. *J Thromb Haemost* ;2020; [epub ahead of print]. doi: 10.1111/jth.14867

518 50. Xiao M, Zhang Y, Zhang S, Qin X, Xia P, Cao W et al. Brief report: Anti-phospholipid  
519 antibodies in critically ill patients with coronavirus disease 2019 (COVID-19) . *Arthritis*  
520 *Rheumatol* 2020; [Epub ahead of print]. doi:10.1002/art.41425

521 51. Amezcua-Guerra LM, Rojas-Velasco G, Brianza-Padilla M, Vazquez-Rangel A,  
522 Marquez-Velasco R, Baranda-Tovar F, et al. Presence of antiphospholipid antibodies in  
523 COVID-19: a case series study . *Ann Rheum Dis* 2020; published online. doi:  
524 10.1136/annrheumdis-2020-218100

525 52. Devreese KMJ, Linskens EA, Benoit D, Peperstraete H. Antiphospholipid antibodies in  
526 patients with COVID-19: a relevant observation? *J Thromb Haemost* 2020; **18**: 2191-2201

527 53. Bertin D, Brodovitch A, Beziane A, Hug S, Bouamri A, Mege JL, Bardin N. Anti-  
528 cardiolipin IgG autoantibodies are an independent risk factor of COVID-19 severity.  
529 *Arthritis Rheumatol* 2020; [epub ahead of print]. doi: 10.1002/art.41409

530 54. Helms J, Tacquard C, Severac F, Leonard-Lorant I, Ohana M, Delabranche X, et al. High  
531 risk of thrombosis in patients with severe SARS-CoV-2 infection: a multicenter prospective  
532 cohort study. *Intensive Care Med* 2020; **4**: 1-10

533 55. Zuo Y, Estes SK, Gandhi AA, Yalavarthi S, Ali RA, Shi H, et al. Prothrombotic  
534 antiphospholipid antibodies in COVID-19. MedRxiv (2020b) , preprint not peer-reviewed.  
535 doi: 10.1101/2020.06.15.20131607

536 56. Vitte J, Bailo Diallo A, Boumaza A, Lopez A, Michel M, Allardet-Servent J, Mezouar S,  
537 et al. A granulocytic signature identifies COVID-19 and its severity. *J Infect Dis* 2020b; in  
538 press, JID-70456R1

539 57. Connors JM, Levy JH. COVID-19 and its implications for thrombosis and  
540 anticoagulation. *Blood* 2020; **135**(23). [epub ahead of print]. doi: 10.1182/blood.2020006000

541 58. Lippi G, Favaloro EJ. D-dimer is Associated with Severity of Coronavirus Disease 2019:  
542 A Pooled Analysis. *Thromb Haemost* 2020; **120**(5): 876-878

543 59. Sakka M, Connors JM, Hékimian G, Martin-Toutain I, Crichi B, Colmegna I, et al.,  
544 Association between D-dimer levels and mortality in patients with coronavirus disease 2019  
545 (COVID-19: a systematic review and pooled analysis. *J Med Vasc* 2020; **45**(5): 268-274

546 60. Zhang L, Yan X, Fan Q, Liu H, Liu X, Liu Z, Zhang Z. D-dimer levels on admission to  
547 predict in-hospital mortality in patients with Covid-19. *J Thrombos Heamost* 2020; **18**(6):  
548 1324-1329

549 61. Leonard-Lorant I, Delabranche X, Severac F, Helms J, Pauzet C, Collange O, et al. Acute  
550 Pulmonary Embolism in COVID-19 Patients on CT Angiography and Relationship to D-  
551 Dimer Levels. *Radiology* 2020; [epub ahead of print]. doi:10.1148/radiol.2020201561

552 62. Faggiano P, Bonelli A, Paris S, Milesi G, Bisegna S, Bernardi N et al. Acute pulmonary  
553 embolism in COVID-19 disease: Preliminary report on seven patients. *Int J Cardiol* 2020;  
554 **313**. [epub ahead of print]. doi: 10.1016/j.ijcard.2020.04.028

555 63. Zhang C, Shen L, Le KJ, Pan MM, Kong LC, Gu ZC, et al. Thromboembolism in  
556 Hospitalized Coronavirus Disease 2019 Patients: A Systematic Review and Meta-Analysis.  
557 *Front Cardiovascular Med* 2020; **7**: article 151 2020

558 64. Poissy J, Goutay J, Caplan M, Parmentier E, Duburcq T, Lassale F, et al. Pulmonary  
559 embolism in patients with COVID-19. *Circulation* 2020; **142**:184-186

560 65. Klok FA, Kruip MJHA, van der Meer NJM, Arbous MS, Gommers DAMPJ, Kant KM, et  
561 al. Incidence of thrombotic complications in critically ill ICU patients with COVID-19.  
562 *Thromb Res* 2020; **191**: 145-147

563 66. Majidi S, Fifi JT, Ladner TR, Lara-Reyna J, Yaeger KA; Yim B, et al. Emergent large  
564 vessel occlusion stroke during New York city's COVID-19 outbreak. *Stroke* 2020; **51**(9):  
565 2656-2663

566 67. Oxley TJ, Mocco J, Majidi S, Kellner CP, Shoirah H, Singh IP et al. Large-vessel stroke  
567 as a presenting feature of Covid-19 in young. *N Engl J Med* 2020; **382**:e60

568 68. Campbell CM, Kahwash R. Will complement inhibition be the new target in treating  
569 COVID-19 related systemic thrombosis ? *Circulation* 2020; **141**: 1739-1741 .

570 69. Merrill JT, Erkan D, Winakur J, James JA. Emerging evidence of a COVID-19 thrombotic  
571 syndrome has treatment implications . *Nature Rev Rheum* 2020; [epub ahead of print]. doi:  
572 10.1038/s41584-020-0474-5

573 70. Carsana L, Sanzagni A, Nasr A, Rossi RS, Pellegrinelli A, Zerbi P, et al. (2020).  
574 Pulmonary post-mortem findings in a series of COVID-19 cases from northern Italy: a two-  
575 centre descriptive study. *Lancet* 2020; published online. doi:10.1016/S1473-3099(20)30434-5

576 71. Marietta M, Coluccio V, Luppi M. COVID-19, coagulopathy and venous  
577 thromboembolism: more questions than answers. *Int Emerg Med* 2020; **11**: 1-13

578 72. Su H, Yang M, Wan C, Yi LX, Tang F, Zhu HY, et al. Renal histopathological analysis of  
579 26 postmortem findings of patients with COVID-19 in China. *Kidney Int* 2020; **98**: 219–227

580 73. Goshua G, Pine AB, Meizlish ML, Chang CH, Zhang H, Bahel P, et al. Endotheliopathy  
581 in COVID-19-associated coagulopathy: evidence from a single-centre, cross-sectional study.  
582 *Lancet Haematol* 2020. [epub ahead of print]. doi: 10.1016/S2352-3026(20)30216-30217

583 74. Libby P, Lüscher T. COVID-19 is, in the end, an endothelial disease. *Europ Heart J* 2020;  
584 **4**: 3038-3044

585 75. O'Sullivan JM, Mc Gonagle D, Ward SE, Preston RJS, O'Donnell JS. *Lancet* 2020; **7**:  
586 e553-e555

587 76. Magro C, Mulvey JJ, Berlin D, Nuovo G, Salvatore S, Harp J, Baxter-Stoltzfus A,  
588 Laurence J. Complement associated microvascular injury and thrombosis in the pathogenesis  
589 of severe COVID-19 infection: a report of five cases. *Transl Res* 2020; **220**: 1-13

590 77. Siguret V, Voicu S, Neuwirth M, Delrue M, Gayat E, Stépanian A, Mégarbane B. Are  
591 antiphospholipid antibodies associated with thrombotic complications in critically ill COVID-  
592 19 patients ? *Thromb Res* 2020; **195**:74-76

593 78. Borghi MO, Beltagy A, Garrafa E, Curreli D, Cecchini G, Bodio C, et al. Anti-  
594 phospholipid antibodies in COVID-19 are different from those detectable in the anti-

phospholipid syndrome. *MedRxiv* Preprint July 16, 2020, not peer-reviewed.  
doi:10.1101/2020.06.17.20134114

79. Cavalli E, Bramanti A, Ciurleo R, Tchorbanov AI, Giordano A, Fagone P et al. Entangling COVID-19 associated thrombosis into a secondary antiphospholipid antibody syndrome: diagnostic and therapeutic perspectives (Review). *Int J Mol Med* 2020; **46**: 903-912

80. Cervera R, Serrano R, Pons-Estel GJ, Ceberio-Hualde L, Shoenfeld Y, et al. Morbidity and mortality in the antiphospholipid syndrome during a 10-year period: a multicentre prospective study of 1000 patients . *Ann Rheum Dis* 2015; **74**: 1011- 1018

81. Martirosyan A, Aminov R, Manukyan G. Environmental triggers of autoreactive responses: induction of antiphospholipid antibody formation. *Front Immunol* 2019; **10**: 1609

82. Catoggio C, Alvarez-Uria A, Fernandez PL, Cervera R, Espinosa G. Catastrophic antiphospholipid syndrome triggered by fulminant disseminated herpes simplex infection in a patient with systemic lupus erythematosus. *Lupus* 2012; **21**: 1359-1361

83. Mendoza-Pinto C, Garcia-Carrasco M, Cervera R. Role of infectious disease in the antiphospholipid syndrome (including its catastrophic variant). *Curr Rheumatol Rep* 2018; **20**: 62

84. Giron-Gonzalez JA, Garcia del Rio E, Rodriguez C, Rodríguez- Martorell J, Serrano A. Antiphospholipid syndrome and asymptomatic carriers of antiphospholipid antibody: prospective analysis of 404 individuals. *J Rheumatol* 2004; **31**:1560–1567

85. Ruiz-Irastorza G, Egurbide MV, Ugalde J. et al.High impact of antiphospholipid syndrome on irreversible organ damage and survival of patients with systemic lupus erythematosus. *Arch Intern Med* 2004; **164**(1): 77-82

86. Ruiz-Irastorza G, Cuadrado MJ, Ruiz-Arruza I, Brey R, Crowther M, R Derksen, et al. Evidence-based recommendations for the prevention and long-term management of thrombosis in antiphospholipid antibody-positive patients: Report of a Task Force at the 13th International Congress on Antiphospholipid Antibodies<sup>[SEP]</sup> *Lupus* 2011; **20**: 206

87. Cohen H, Hunt BJ, Efthymiou M, Arachchillage DRJ, Mackie IJ, Clawson S, et al. Rivaroxaban versus warfarin to treat patients with thrombotic antiphospholipid syndrome, with or without systemic lupus erythematosus (RAPS): a randomised, controlled, open-label, phase 2/3, non-inferiority trial. *Lancet Haematol* 2016; **3**: e426–e436

88. Resseguier AS, Pereira B, Rieu V, Le Guenno G, Grobost V, Ruivard M. Direct oral anticoagulants: an alternative treatment for thrombotic antiphospholipid syndrome? *Lupus* 2017; **26**: 1297–1303.

89. Pengo V, Denas G, Zoppellaro G, Jose SP, Hoxha A, Ruffatti A, et al. Rivaroxaban vs warfarin in high-risk patients with antiphospholipid syndrome. *Blood* 2018; **132**: 1365–1371

631 90. Tektonidou MG, Andreoli L, Limper M, Amoura Z, Cervera R, Costedoat-Chalumeau N,  
632 et al. EULAR recommendations for the management of antiphospholipid syndrom in adults.  
633 *Ann Rheum Dis* 2019; **78**: 1296-1304

634 91. Belizna C. Hydroxychloroquine as an anti-thrombotic in antiphospholipid syndrome.  
635 *Autoimmun Rev* 2015; **14**: 358-362

636 92. Nuri, E, Taraborelli, M, Andreoli, L, Tonello M, Gerosa M, Calligaro A, et al. Long-term  
637 use of hydroxychloroquine reduces antiphospholipid antibodies levels in patients with  
638 primary antiphospholipid syndrome. *Immunol Res* 2017; **65**: 17–24.

639 93. Wang TF, Lim W. What is the role of hydroxychloroquine in reducing thrombotic risk in  
640 patients with antiphospholipid antibodies? *Hematology* 2016; **1**: 714-716

641 94. Pengo V, Ruffatti A, Legnani C, Gresele P, Barcellona D, Erba N, Testa S, Marongiu F,  
642 Bison E, Denas G, Banzato A, Padayattil JS, Lliceto S. Clinical course of high-risk patients  
643 diagnosed with antiphospholipid syndrome. *J Thromb Haemost* 2010; **8**: 237-242

644 95. Wallace DJ. Does hydroxychloroquine sulfate prevent clot formation in systemic lupus  
645 erythematosus ? *Arthritis Rheum* 1987; **30**: 1435-1436

646 96. Loudon JR. Hydroxychloroquine and postoperative thromboembolism after total hip  
647 replacement. *Am J Med* 1988; **85**: 57-61.

648 97. Jancinova V, Nosal R, Petrikova M. On the inhibitory effect of chloroquine on blood  
649 platelet aggregation. *Thromb Res* 1994; **74**: 495-504

650 98. Edwards MH, Pierangeli S, Liu X, Barker JH, Anderson G, Harris EN.  
651 Hydroxychloroquine reverses thrombogenic properties of antiphospholipid antibodies in mice.  
652 *Circulation* 1997; **96**: 4380-4384

653 99. Miranda S, Billoir P, Damian L, Thiebault PA, Schapman D, Le Besnerais M, et al.  
654 Hydroxychloroquine reverses the prothrombotic state in a mouse model of antiphospholipid  
655 syndrome: role of reduced inflammation and endothelial dysfunction. *PLoS One* 2019; **14**(3):  
656 e0212614

657 100. Erkan D, Yazici Y, Peterson MG, Sammaritano L, Lockshin MD. A cross-sectional  
658 study of clinical thrombotic risk factors and preventive treatments in antiphospholipid  
659 syndrome. *Rheumatology* 2002; **41**: 924-929

660 101. Ho KT, Ahn CW, Alarco GS, Baethge BA, Tan FK, Roseman J, et al. Systemic lupus  
661 erythematosus in a multicentric cohort (LUMINA): XXVIII. Factor predictive of thrombotic  
662 event. *Rheumatology* 2005; **44**: 1303-1307

663 102. Erkan D, Lockshin. APS ACTION-AntiPhospholipid Syndrome Alliance for Clinical  
664 Trials and InternatiOnal Network . *Lupus* 2012; **21**: 695-698

665 103. Erkan D, Unlu O, Sciascia S, Belmont HM, Ware Branch D, Cuadrado MJ, et al.  
666 Hydroxychloroquine in the primary thrombosis prophylaxis of antiphospholipid antibody  
667 positive patients without systemic autoimmune disease. *Lupus* 2018; **27**: 399-406

668 104. Metjian A, Lim W. ASH evidence-based guidelines: should asymptomatic patients with  
669 antiphospholipid antibodies receive primary prophylaxis to prevent thrombosis? *Hematology*  
670 *Am Soc Hematol Educ Program* 2009; 247-249.

671 105. Ruiz-Irastorza G, Egurbide MV, Pijoan JL, Garmendia M, Villar I, Martinez-Berriotxo  
672 A. et al. Effect of antimalarials on thrombosis and survival in patients with systemic lupus  
673 erythematosus. *Lupus* 2006; **15**: 577-583

674 106. Jung H, Bobba R, Su J, Shariati-Sarabi Z, Gladman DD, Urowitz M, et al. The protective  
675 effect of antimalarial drug on thrombovascular events in systemic lupus erythematosus.  
676 *Arthritis Rheum* 2010; **62**: 863-868

677 107. Espinola RG, Pierangeli SS, Gharavi AE, Harris EN. Hydroxychloroquine reverse  
678 platelet activation induced by human IgG antiphospholipid antibodies. *Thromb Haemost*  
679 2002; **87**: 518-522.

680 108. Rand JH, Wu XX, Quinn AS, Chen PP, Hathcock JJ, Taatjes DJ. Hydroxychloroquine  
681 directly reduces the binding of antiphospholipid antibody-beta2-glycoprotein I complexes to  
682 phospholipid bilayers. *Blood* 2008; **112**: 1687-1695

683 109. Rand JH, Wu XX, Quinn AS, Ashton AW, Chen PP, Hathcock JJ, Andree HA, Taatjes  
684 DJ. Hydroxychloroquine protect the annexin A5 anticoagulant shield from disruption by  
685 antiphospholipid antibodies: evidence for a novel effect for an old anti-malarial drug. *Blood*  
686 2010; **115**: 2292-2299

687 110. Petri M. Use of hydroxychloroquine to prevent thrombosis in systemic lupus  
688 erythematosus and in antiphospholipid antibody-positive patients. *Curr Rheumatol Rep* 2011;  
689 **13**: 77-80.

690 111. Al-Homood IA. Thrombosis in systemic lupus erythematosus: a review article. *ISRN*  
691 *Rheumatol* 2012; **2012**:428269

692 112. Schmidt-Tanguy A, Voswinkel J, Henrion D, Subra JF, Loufrani LL, Rohmer V, Ifrah N,  
693 Belizna C. Antithrombotic effects of hydroxychloroquine in primary antiphospholipid  
694 syndrome patients. *Int Soc Thromb Haemost* 2013; **11**(10): 1927-1929

695 113. Albert CR, Schlesinger WJ, Viall CA, Mulla MJ, Brosens JJ, Chamley LW, Abrahams  
696 VM. Effect of hydroxychloroquine on antiphospholipid antibody-induced changes in first  
697 trimester trophoblast function. *Am J Reprod Immunol* 2014; **71**(2): 154-164

698 114. Ruffatti A, Tonello M, Hoxha A, Sciascia S, Cuadrado MJ, Latino JO, et al. Effect of  
699 additional treatments combined with conventional therapies in pregnant patients with high-  
700 risk antiphospholipid syndrome: a multicentre study. *Thromb Haemost* 2018; **118**: 639-648

115. Schreiber K, Breen K, Cohen H, Jacobsen S, Middeldorp S, Pavord S, et al. Hydroxychloroquine to improve Pregnancy outcome in women with Antiphospholipid Antibodies (HYPATIA) protocol: a multinational randomized controlled trial of hydroxychloroquine versus placebo in addition to standard treatment in pregnant women with antiphospholipid syndrome or antibodies. *Semin Thromb Hemost* 2017; **43**: 562–571
116. Belizna C, Pregnolato F, Abad S, Alijotas-Reig J, Amital H, Amoura Z, et al. HIBISCUS: hydroxychloroquine for the secondary prevention of thrombotic and obstetrical events in primary antiphospholipid syndrome. *Autoimmun Rev* 2018; **17**: 1153–1168.
117. Mangolin S, Mazetto BM, Annichino-Bizzacchi JM, Andrade Orsi F. Study of the role of hydroxychloroquine in immunomodulation and control of hypercoagulability in patients diagnosed with primary antiphospholipid syndrome. *Blood* 2019; **134**: 3682
118. Kravvariti E, Koutsogianni A, Samoli E, Sfrikakis PP, Tektonidou MG. The effect of hydroxychloroquine on thrombosis prevention and antiphospholipid syndrome: a pilot open label randomized prospective study. *Autoimmun Rev* 2020; **19**(4):102491
119. Pal A, Pawar A, Goswami K, Sharma P, Prasad R. Hydroxychloroquine and COVID-19: a cellular and molecular biology based update. *Indian J Clin Biochem* 2020; **35**: 274-284
120. Connell NT, Battinelli EM, Connors JM. Coagulopathy of COVID-19 and antiphospholipid antibodies. *J Thromb Haemost* (2020) 00: 1-2
121. Mathian A, Mahevas M, Rohmer J, Roumier M, Cohen-Aubart F, Amador-Borrero B, et al. Clinical course of coronavirus disease 2019 (COVID-19) in a series of 17 patients with systematic lupus erythematosus under long-term treatment with hydroxychloroquine. *Ann Rheum Dis* 2020; **79**(6): 837-839
122. Gautret P, Million M, Kaplanski G, Camoin-Jau L, Colson P, Fenollar F. et al. Natural history of COVID-19 and therapeutic options. *Exp Rev Clin Immunol* 2020. in press.