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Brief Reviews

The Journal of Immunology

COVID-19 as an Acute Inflammatory Disease

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The 2019 coronavirus disease (COVID-19) pandemic caused by the virus severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) has created an unprecedented global crisis for the infrastructure sectors, including economic, political, healthcare, education, and research systems. Although over 90% of infected individuals are asymptomatic or manifest noncritical symptoms and will recover from the infection, those individuals presenting with critical symptoms are in urgent need of effective treatment options. Emerging data related to mechanism of severity and potential therapies for patients presenting with severe symptoms are scattered and therefore require a comprehensive analysis to focus research on developing effective therapeutics. A critical literature review suggests that the severity of SARS-CoV-2 infection is associated with dysregulation of inflammatory immune responses, which in turn inhibits the development of protective immunity to the infection. Therefore, the use of therapeutics that modulate inflammation without compromising the adaptive immune response could be the most effective therapeutic strategy. ***The Journal of Immunology*****, 2020, 205: 000–000.**

Following reports of patients with severe pneumonia caused by a β coronavirus in China, the World Health Organization (WHO) named the causative agent severe acute respiratory syndrome (SARS) coronavirus (SARS-CoV)-2 and named the disease as the 2019 novel coronavirus disease (COVID-19). There is a high homology between SARS-CoV-2 and SARS-CoV, as well as the Middle East respiratory syndrome (MERS) coronavirus (MERS-CoV) (1). The clinical manifestations of COVID-19 include asymptomatic carriers, presymptomatic carriers, and symptomatic patients with acute respiratory distress syndrome (ARDS) or pneumonia (2, 3). Although the incubation period for COVID-19 varies between 4 and 14 days, one study reported that over 97% of infected individuals who were presymptomatic developed clinical symptoms within 11–12 days (4). The prevalence of asymptomatic cases is over 80%, and cases are defined as individuals with positive viral tests but without any COVID-19 symptoms (2, 5, 6). Among symptomatic patients, the severity of illness ranges from mild to moderate pneumonia symptoms (fever, fatigue, and cough) (81%), severe pneumonia symptoms (dyspnea, tachypnea with respiratory rates ≥30/min, and hypoxia) and lung infiltrates (14%), and critical condition associated with respiratory failure or multiorgan system dysfunction (5%) (7). The most serious complications of COVID-19 are sepsis-like inflammation, coagulopathy, and respiratory or cardiovascular complications. In response to injury or infection, the innate immune system mounts immediate inflammatory responses to limit the infection and to help the adaptive immune system develop long-lasting, host-protective Abs and T cell responses against the virus within 7–10 days postinfection. However, when inflammation is not modulated or resolved after serving its purpose, it turns into hyperinflammation or becomes chronic and results in the inhibition of adaptive immune responses, tissue damage, or organ failure. Such dysregulated inflammation results in a "cytokine storm" that is evident in sepsis as well as in patients with severe respiratory diseases caused by coronaviruses such as SARS, MERS, and COVID-19 (8, 9). A cytokine storm is manifested by uncontrolled production of inflammatory cytokines such as IL-6, G-CSF, IP-10, MCP-1, MIP-1α, TNF-α, IL-10, IL-7, and IL-2, which are significantly higher in intensive care unit (ICU) patients than non-ICU patients hospitalized with COVID-19 (10). A cytokine storm causes lymphopenia and prevents the adaptive immune system to produce antiviral Abs. Emerging evidence suggest that complications of COVID-19 are associated with a gender or age disparity in inflammatory immune responses to SARS-CoV-2 infection as well as underlying health issues. Therefore, understanding and successfully controlling inflammation would be a promising approach for the management of COVID-19, as discussed below.

The severity of COVID-19 is associated with a gender, age, or health disparity in the immune response: inflammation

Emerging evidence suggests a higher rate of death in men compare with women who are infected with SARS-CoV-2 (11).

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Abbreviations used in this article: ACE2, angiotensin-converting enzyme II; ARDS, acute respiratory distress syndrome; COVID-19, 2019 novel coronavirus disease; COX, cyclooxygenase; CQ, chloroquine; HCQ, hydroxychloroquine; ICU, intensive care unit; MERS, Middle East respiratory syndrome; MERS-CoV, Middle East respiratory syndrome coronavirus; NSAID, nonsteroidal anti-inflammatory drug; SARS, severe acute respiratory syndrome; SARS-CoV, severe acute respiratory syndrome coronavirus; WHO, World Health Organization.

As of May 6, 2020, mortality of men exceeded that of women worldwide (Fig. 1, $p < 0.000001$). The gender-related susceptibility to COVID-19 is due to sex differences in innate as well as adaptive immunity. Similar to COVID-19, in acute inflammatory sepsis, women survive better than men (12). Although women and men mount inflammatory immune responses to pathogens, women resolve acute inflammation and prevent hyperinflammation better than men (13). Such ability of women to modulate inflammation without compromising adaptive immune responses is in part due to higher production of specialized proresolving mediators such as lipoxins, protectins, resolvins, and maresins (13). The ability of women in modulating inflammation has been shown by inducing PBMCs by various ligands for the innate TLR, TLR2, TLR4, or TLR7/8, which resulted in the release of higher levels of inflammatory cytokines in men compared with women (14). The X chromosome perhaps plays a key role in the induction and resolution of inflammation because many proteins that are involved in immune responses are encoded on the X chromosome (15). For instance, TLRs, CD40L, and the main proteins associated with NF-κB signaling pathway are linked to the X chromosome (16). Although one of the two X chromosomes in females is randomly inactivated by methylation, ~15% of X-linked genes escape this process of methylation, thereby increasing the X-linked proteins in women compared with men (17, 18). In fact, females are composed of a mosaic of cells from paternal and maternal X chromosomes, providing them with greater diversity of immune responses (16, 19) and enabling them to show lower levels of some inflammatory cytokines and better T cell and Ab responses compared with men (20). Given that inflammatory markers are significantly different between prepubertal boys and girls, sex chromosomes appear to be more important than sex hormones during inflammation (21). This notion has been supported by data from X-linked diseases. In subjects with Turner syndrome, who are phenotypically female but carry one X chromosome, inflammatory responses are similar to males (22). In subjects with Klinefelter syndrome who are phenotypically male but carry two X chromosomes like females, inflammatory responses are similar to females (14). This is despite a higher level of testosterone in individuals with Klinefelter syndrome than in women. These data suggest that X chromosome mosaicism on X-linked genes is involved in the TLR signaling pathways. Perhaps, the lower secretion of inflammatory cytokines in women as well as their ability to resolve inflammation could protect them from life-threatening inflammatory responses during sepsis, trauma, or COVID-19.

Molecular polymorphism in the innate and adaptive immune systems reflected in the TLR and HLA systems, respectively, could explain gender disparity associated with COVID-19 symptoms. Both SARS-CoV and SARS-CoV-2 are pH dependent and require acidification of endosome (23, 24) as well as lysosome (25) for infecting the cells. Similar to SARS-CoV, ssRNA of SARS-CoV-2 will likely bind TLR3/7/8 in the endosome and lysosome, resulting in the induction of innate inflammatory responses such as type I IFNs (26), which are involved in viral clearance. A greater expression of TLR7 in women compared with men (27) could help them to better cope with COVID-19 by producing highly tailored but transient antiviral inflammatory

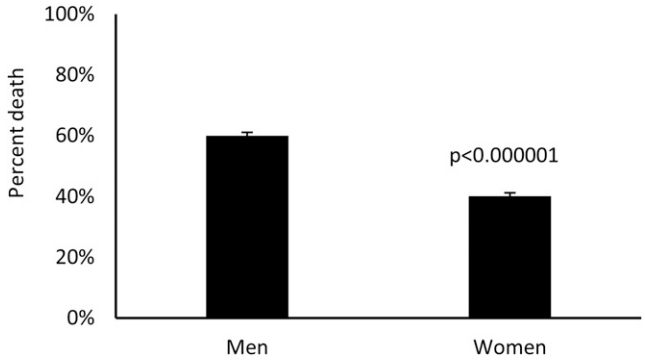

FIGURE 1. Gender-disaggregated cases of death. Average of COVID-19 fatality in men and women reported by the Global Health 5050 website, http://globalhealth5050.org/covid19/. Graph represents the data from 48 countries as of May 6, 2020. Statistical analysis was performed using two-tailed Student *t* test.

cytokines. Upon stimulation of TLR7 and TLR8, women produce higher amounts of antiviral IFN-α and similar levels of TNF-α, respectively (28, 29). Such gender disparity in TLR activity was reported to be associated with a lower HIV-1 viral load in women compared with men (29). In addition, polymorphism in TLR7 might be involved in susceptibility to SARS-CoV-2 infection associated with severity of the symptoms. To this end, specific nonsynonymous single nucleotide polymorphisms in TLR7 have been reported to be associated with greater susceptibility of males to hepatitis B infection compared with females (30). In hepatitis C infection, specific TLR7/8 polymorphisms are associated with greater antiviral IFN-α and lower levels of proinflammatory cytokines upon stimulation (31). In addition to the polymorphisms in the innate immune response, MHC class II polymorphism may also play a role. Processing of SARS-CoV-2 in the lysosome could makes viral proteins available to MHC class II Ag presentation. This presentation could be influenced by highly polymorphic HLA-DP, -DQ, -DR, and -DM in modulating immune responses, as reported in other inflammatory diseases (32). However, data from population genetic studies in patients with SARS are inconclusive, as some reports show the association of HLA polymorphism with the severity of the infection (33), whereas some other reports show no correlation (34). This could be due to different experimental design, as some correlated the severity of the disease whereas others correlated the disease incidence with HLA polymorphism. It is yet to be determined whether severe symptoms are associated with certain HLA polymorphism while randomizing patients based on gender, age, and underlying diseases.

Mortality of COVID-19 worldwide is also caused by respiratory failure, cardiovascular failure, and multiorgan failure secondary to ARDS, coagulopathy, shock, and arrythmia, especially in adults older than 65 and people with underlying health conditions such as asthma, diabetes mellitus, cardiovascular disease, or cancer (35, 36). It was reported that patients with a history of cancer had higher incidence of severe symptoms or death compared with those who did not have cancer (36). This overall increase in mortality is because patients with these conditions also suffer from chronic inflammation. The impact of age is due to the immune system becoming dysregulated and increased levels of inflammatory cytokines as we age (37). To this end, age does not seem to be an independent risk factor for patients with COVID-19;

rather, it is correlated with gender disparity in inflammatory immune responses. As men and women age, the anti-inflammatory angiotensin-converting enzyme II (ACE2) significantly decreases in men, but it increases in women (38).

Viremia or dysregulated immune responses? A hyperactive inflammatory immune response dismantles adaptive immune responses

Although patients with severe COVID-19 tend to have a high viral load (39), the viral load in asymptomatic patients is similar to that of symptomatic patients (6). These data suggest that viral load may not be the primary cause of fatality in patients with SARS-CoV-2 infection. In contrast, although no elevated levels of inflammatory cytokines/chemokines were reported in asymptomatic patients, inflammatory responses were consistently detected in symptomatic patients. Circulating levels of cytokines (IL-6, TNF-α) and chemokines (CXCL10, CCL2) involved in the cytokine storm syndrome are elevated in COVID-19, which could promote hyperinflammation, leading to ARDS and multiorgan failure (Refs. 40, 41, and S. Wan, Q. Yi, S. Fan, J. Lv, X. Zhang, L. Guo, C. Lang, Q. Xiao, K. Xiao, Z. Yi, et al., manuscript posted on medRxiv). ICU patients with severe disease had higher plasma levels of IL-2, IL-7, IL-10, G-CSF, IP-10, MCP-1α, MIP-1, and TNF-α, again suggesting a cytokine storm associated with severity of the disease (10). These data suggest that mortality due to organ failure might be because of hyperinflammation similar to a cytokine storm seen in sepsis. A cytokine storm and lymphopenia were also evident in patients with SARS and MERS (42–44). This cytokine storm perhaps initiates viral sepsis and inflammatory-induced organ failure. Similar to sepsis, patients with severe COVID-19 manifest inflammatory cytokines associated with lymphopenia, in addition to decreased antiviral IFN-γ production by $CD4^+$ T cells (45). This suggests that hyperinflammation prevents the establishment of antiviral adaptive immune responses for the clearance of the virus. In fact, patients with severe symptoms failed to clear the virus, whereas patients with mild symptoms were able to develop immunity and clear the virus (39). Inflammatory cytokines such as TNF-α and IL-6 could induce apoptosis in lymphocytes, causing lymphopenia (46). High levels of IL-2 could also promotes activation-induce cell death in lymphocytes (47). In addition, lymphocytes express the coronavirus receptor ACE2 and may be a direct target of COVID-19–induced apoptosis (48). Also, patients with severe symptoms have elevated lactic acid levels in the blood, which might suppress the proliferation of lymphocytes (49).

SARS-CoV-2 can trigger innate inflammatory responses via several pathways. One pathway involves TLRs. As a ssRNA viruses, SARS-CoV and SARS-CoV-2 invade the cells using endosomal pathway and releases genomic RNA into the endosome (23, 24) to bind TLR7/8 and trigger inflammatory responses in the lungs. ACE2 also contributes to inflammation in the lungs. ACE2 is the cellular receptor for spike (S) protein of SARS-CoV and SARS-CoV-2 (50–52), with 10- to 20-fold higher affinity to SARS-CoV-2 than to SARS-CoV (53). ACE2 is mainly expressed in type 2 alveolar cells in the lungs, myocardial cells, kidney proximal tubule cells, bladder urothelial cells, and testis (54–56). TLR7 and TLR8 are also expressed in the lungs (57, 58). These are the organs that are involved in severe COVID-19. Physiological activity of ACE2 is vital to control inflammation in the lungs by hydrolyzing the inflammatory angiotensin II to anti-inflammatory angiotensin 1–7 (59). In patients with severe clinical symptoms, ACE2 is depleted by SARS-CoV-2 infection (60). Reduction of ACE2 can cause dysfunction of the renin–angiotensin system and enhance inflammation and pulmonary edema through an increase in angiotensin II levels (61). Angiotensin II induces several inflammatory responses by signaling through AT1R (62, 63) and upregulation of E-selectin, P-selectin, IL-8, CCL5, and CCL2 (MCP1) expression in endothelial cells (62, 64). Angiotensin II can also induce TLR4 activation triggering the innate immune response (65). In addition, depletion of ACE2 decreases the production of angiotensin 1–7, which has an anti-inflammatory and antifibrotic activity (60). This facilitates massive inflammatory responses in the lungs.

Potential therapeutics for the management of patients with severe COVID-19

Some therapeutics are mainly focused on the control of viremia for the management of COVID-19 patients who manifest severe symptoms. Also, there are some controversial reports (T. Siekmann and K.T. Kopec, manuscript posted on emDocs) on the efficacy of corticosteroids for the control of hyperinflammation in patients with COVID-19. To this end, emerging evidence suggests that nonsteroidal drugs that reduce inflammation and modulate the innate immune response by inhibiting a cytokine storm without compromising the adaptive immune response could be more effective for the management of patients with severe symptoms. This is because adaptive arms of the immune system including antiviral Ab production in the presence of help from $CD4^+$ T cells, as well as $CD8^+$ T cell responses, are required for clearance of the virus and establishment of immunological memory to protect the host from a recall infection. It takes 7–10 d for an adaptive immune response to be established following viral infection. In the case of SARS, antiviral memory $CD8^+$ T cells as well as neutralizing Ab response to SARS-CoV showed long-lasting immunity that protected the host from recall infection (66, 67).

Control of viremia by antiviral therapies may not be the best therapeutic strategy. Chloroquine (CQ) or its less toxic metabolite hydroxychloroquine (HCQ) is suggested to inhibit cellular processing of SARS-CoV-2 in vitro (24). CQ is an antimalarial drug that blocks autophagy by reducing acidity of endosome and lysosome and preventing the virus–cell fusion; it also interferes with glycosylation of the ACE2 receptor and inhibit the binding of SARS-CoV to ACE2 (68), although such mechanism has not been shown for SARS-CoV-2. Studies in cell culture suggested that CQ can cripple the virus, but the doses needed are usually high, which could cause severe toxicities such as cardiovascular effects (arrythmia and cardiomyopathy resulting in cardiac failure, sometimes fatal), hematologic effects (bone marrow suppression), and hypoglycemia. In patients with chronic diseases, who often show severe symptoms, both CQ and HCQ cause severe hypoglycemia (69). In addition, when used in patients with glucose-6-phosphate dehydrogenase (G6PD) deficiency at higher than normal therapeutic doses, there is a high risk for hemolytic anemia (70). Importantly, both CQ and HCQ are metabolized by hepatic cytochrome P450 enzyme 2D6 (CYP2D6), which is genetically polymorphic

among individuals (71). CYP2D6 polymorphisms lead to a wide variation in blood HCQ concentrations, thus rendering the drug either ineffective or toxic in patients with CYP2D6 polymorphisms (71). Approximately 7% of white Americans have no functional CYP2D6, which could increase the toxicity of the drug (72). Such a genetic variability influences the response to treatment and increases the risk of toxicity (73). Chinese experts recommended a twice daily use of CQ phosphate tablet (500 mg) for 10 d for patients with symptomatic COVID-19 pneumonia and without contraindications to CQ (74). However, results on the efficacy of CQ or HCQ against COVID-19, in vivo, are murky. By referring to a Chinese Clinical Trial Registry, a letter to *Bioscience Trends* (75) claims that results from more than 100 patients showed CQ phosphate was superior to the control treatment in inhibiting the progression of pneumonia and reducing SARS-CoV-2 viral load, but without publishing data. Other COVID-19 studies in China using CQ or HCQ have not been shared with WHO (76). A recent clinical trial approved by the French Ministry of Health reported that use of 200 mg of HCQ sulfate three times daily alone (14 patients) or with azithromycin (six patients) for 10 d reduced viral load in nasopharyngeal swabs (77), but the trial was not randomized. Treated patients who were asymptomatic accounted for 10% (2/20), but symptomatic patients were not randomized for those with upper respiratory tract infection showing rhinitis, pharyngitis, or isolated low-grade fever and myalgia, nor were patients with lower respiratory tract infections with symptoms of pneumonia or bronchitis. In addition, clinical outcomes such as deaths were not reported. A retrospective analysis of data from 368 patients with COVID-19 hospitalized in the United States Veterans Health Administration medical centers showed that taking HCQ alone or in combination with azithromycin did not reduce the risk of mechanical ventilation and that the risk of death was higher in the HCQ group compared with control group (Magagnoli, Narendran, Pereira, Cummings, Hardin, Sutton, and Ambati, manuscript posted on medRxiv). Following this report, the U.S. Food and Drug Administration issued warning about use of HCQ for COVID-19 (https://time.com/5827085/fda-warning-hydroxychloroquine/). A comprehensive review of literature show insufficient evidence on the efficacy of CQ or HCQ against COVID-19 (78). Although CQ or HCQ have anti-inflammatory effects through the inhibition of TLR signaling (TLR7/8/9) (79), CQ directly suppresses proliferation, metabolic activity, and cytokine secretion of human $CD4^+$ T cells by modulating AP-1 signaling (80). This could in turn inhibit antiviral Ab production and adaptive immunity against SARS-CoV-2. This is likely because of the inhibition of protease activity of the lysosome, a cellular compartment involved in Ag processing for MHC class II presentation and activation of $CD4^+$ T cells to assist Ab production by B cells. Direct immune suppressive effects of CQ on B cell activation has also been reported to be through inhibiting Ca^{2+} permeable IP_3R and TRPC3 and/or STIM/Orai channels in B cells (81). Use of CQ and HCQ as part of the standard treatment for patients with autoimmune rheumatoid arthritis and systemic lupus erythematosus is because of their inhibitory effects on the adaptive immune system, which cannot be translated to viral infections in which an adaptive immune response is needed for clearance of the infection. Because CQ and HCQ have prolonged half-lives, their negative impact on the adaptive immune response should be considered (82).

Remdesivir is an investigational antiviral compound that is a nucleoside analog developed by Gilead Sciences to fight Ebola by inhibiting the RNA polymerase to dismantle viral replication. Remdesivir did not help patients with Ebola during the 2019 outbreak in the Democratic Republic of Congo (83), and in a phase II clinical trial, the efficacy of remdesivir was significantly worse than that of the two mAbs MAb114 and REGN-EB3 arms (84). This drug has been considered for patients with COVID-19. A recent study showed that remdesivir can inhibit SARS-CoV-2 in a human liver cancer cell line in vitro (24). Thus far, the use of remdesivir in COVID-19 patients in the United States and Europe has produced anecdotal evidence of benefit. A daily i.v. administration of remdesivir in patients with severe COVID-19 who were hospitalized in the US, Europe, or Japan showed that of patients receiving mechanical ventilation while on remdesivir therapy, 57% were extubated within 18 d follow-up (85). The study was not randomized, and thus it does not show if patients who were extubated were responding to remdesivir, not having other comorbidities, or not being in a high-risk category (men or elderly) compared with those who were not extubated while receiving remdesivir. Another study on patients with severe COVID-19, of which 63% were men and 58% had diabetes mellitus, showed that 33% of patients were extubated within 14 d follow-up without remdesivir, and a greater percentage of patients who died were over 65 y of age (62 versus 37%) (86). Adjusting the results for age, gender, and underlying diseases would determine what percentage of patients who are in a high-risk category were extubated because of remdesivir in one study compared with 33% who were extubated without this medication in another study.

Control of inflammation could be a promising approach for the management of COVID-19. Therapeutic strategies that target the virus rather than the inflammatory immune response have not produced consistent results. Although controlling tissue-damaging hyperinflammation could be a promising approach, highly tailored use of anti-inflammatory compounds that modulate inflammation without compromising the adaptive immune response should be considered. Current results from the use of different inflammatory compounds and potential candidates for the management of patients with severe COVID-19 are evaluated below.

Corticosteroids. Because of the strong correlation between severity of symptoms in patients with COVID-19 and inflammation, anti-inflammatory steroids are suggested for the management of the disease (41). Corticosteroids have been used during the outbreaks of SARS-CoV (87) and MERS-CoV (88) and are being used in combination with other medications in patients with SARS-CoV-2 infection (10). The results in patients with SARS and MERS suggest that corticosteroids not only failed to reduce mortality but also delayed viral clearance (88). Corticosteroid treatment in influenza was even associated with increased mortality (89). A recent report on the use of corticosteroid (40 mg methylprednisolone once or twice per day) in 11 patients with COVID-19 in two hospitals in China showed no efficacy on virus clearance time or duration of symptoms (90). In general, clinical evidence are not supportive of systemic administration of corticosteroids for the

treatment of patients with severe COVID-19 (91); rather, it is more likely that systemic administration of corticosteroids would be harmful because of the suppression of the adaptive immune system. Corticosteroids such as prednisolone act through their nuclear receptors repressing the activity of the transcription factors NF-κB and AP-1, as well as members of the STAT, C/EBP, and NFAT families, thereby having a broad immunosuppressive activity on Ag presentation as well as the suppression of antiviral T cell and Ab responses (92). According to WHO interim guidance, corticosteroids should not be given systemically unless in the setting of a clinical trial (https://www.who.int/publications-detail/clinical-management-of-severe-acute-respiratory-infection-when-novel-coronavirus-(ncov)-infection-is-suspected). Per the Infectious Diseases Society of America guidelines, despite widespread use of corticosteroids during the SARS outbreak, conclusive evidence of benefit was lacking, and administering steroids early in the disease process, before viral replication is controlled, may lead to a delay in viral clearance (https://www.idsociety.org/practice-guideline/covid-19-guideline-treatment-and-management/).

Nonsteroidal anti-inflammatory drugs. Nonsteroidal anti-inflammatory drugs (NSAIDs) such as naproxen and indomethacin have been shown to manifest antiviral activity against SARS-CoV and influenza A and B viruses by inhibiting viral RNA synthesis (93, 94). As SARS-CoV-2 is a ssRNA virus, naproxen is suggested to be effective in patients with COVID-19 because of having both anti-inflammatory and antiviral activity (95). However, their use could also inhibit the induction of an adaptive immune response against the virus. NSAIDs reduce inflammation by inhibiting the cyclooxygenase (COX) enzymes, COX-1 and COX-2, thereby inhibiting the production of PGs and thromboxane A2 (TXA2). The caveat is that COX-2 is also upregulated during activation of human B cells for Ab production. It was reported that ibuprofen, aspirin, naproxen, and acetaminophen inhibit Ab production, with ibuprofen having the greatest inhibitory effect (96). Preclinical studies show that aspirin and ibuprofen can inhibit both MHC class I and MHC class II–restricted Ag presentation in dendritic cells (97). Also, during acute respiratory tract infections, a short-term use of NSAIDs has been reported to be associated with higher rates of complications, including pneumonia (98), which is more likely a complication in patients with severe COVID-19. The implications of these results are that the use of widely available NSAIDs after viral infection or vaccination could alter the ability of the patients to mount antiviral and immune responses.

Highly tailored anti-inflammatory drugs. Coronaviruses that cause severe acute respiratory syndrome (SARS-CoV and SARS-CoV-2) bind to ACE2 in alveoli pulmonis, which then cause lung damage and even lung function failure. Although ACE2 is a receptor for SARS-CoV-2, upon infecting the cells, the virus depletes ACE2, thereby disturbing a normal inflammatory immune response by increasing the inflammatory angiotensin II and decreasing the anti-inflammatory angiotensin 1–7. Therefore, induction of ACE2 is considered as a possible therapeutic strategy for the modulation of inflammation in patients with severe COVID-19 (99). It was suggested that administration of recombinant ACE2 protein can protect the host from severe acute lung injury (100). In Europe, Apeiron Biologics has received approval to test recombinant ACE2 in COVID patients (101). Because there are multiple mechanisms dysregulating inflammatory immune responses, use of recombinant ACE2 alone may not be highly effective, as shown for patients infected with SARS-CoV in 2017 (101).

Statins are shown to be effective inducers of ACE2 allowing patients with Ebola to recover from the infection (102). Also, statins are known inhibitors of the MYD88 pathway (103), which has been shown to be highly activated during infection with SARS-CoV (103). Importantly, they do not significantly alter the expression of MYD88 in normal conditions (103). Angiotensin 1–7 heptapeptide may also be used as a highly tailored anti-inflammatory drug to counteract the activities of angiotensin II when ACE2 is depleted by SARS-CoV-2. Furthermore, drugs such as losartan that block type I angiotensin II receptor could block inflammatory pathways in the lungs without compromising the adaptive immune system (99). Kuba and colleagues (104) found that mice treated with losartan after acid aspiration–induced acute lung injury with the addition of SARS-CoV spike protein had significantly diminished lung injury and pulmonary edema compared with mice treated with placebo. Furthermore, recombinant human ACE2 infusions and losartan both prevented severe lung injury and pulmonary edema in ACE2 depleted mice (104).

Passive immunotherapy by convalescent plasma could be the most promising strategy. Convalescent plasma therapy has been successfully used for the treatment of SARS, MERS, and 2009 H1N1 influenza pandemic with no toxicity (105–107). Although convalescent plasma therapy did not significantly improve the survival patients with Ebola, it could be because of insufficient neutralizing Ab, as Ab titration data were not available (108). Because of the similarity in the virological and clinical characteristics between SARS or MERS and COVID-19, similar efficacy with convalescent plasma therapy is expected for COVID-19. In fact, convalescent plasma containing sufficient neutralizing Ab has improved the clinical outcomes through neutralizing viremia and alleviating inflammation in severe COVID-19 cases (109–111). Because of its efficacy, the U.S. Food and Drug Administration has recommended investigational use of passive immunotherapy by means of convalescent plasma for the treatment of critically ill patients with COVID-19 (112). Because over 80% of infected patients are asymptomatic, they could serve as donors of convalescent plasma for the management of clinically ill patients to prevent or treat severe clinical symptoms, allowing patients develop immunity against the virus.

Conclusions

Emerging data suggest that fatality of COVID-19 is determined by gender, age, or health disparities associated with the innate and adaptive immune responses. To this end, sepsis-like inflammation or a cytokine storm as a result of a hyperactivation of the innate immune system, along with the inhibition of the adaptive immune response, makes COVID-19 deadly for the elderly or individuals with underlying diseases, with men being more vulnerable than women. TLR7/8 polymorphism and/or HLA class II haplotypes could be associated with vulnerability to SARS-CoV-2 infection. Therapeutic strategies for the management of severe symptoms are focused on the control of viremia and/or inflammation. Antiviral therapeutics for alleviating symptoms of the disease

might be effective as a preventive strategy or very early during infection. However, they could prevent the development of protective immunity against the virus, putting patients at risk for recurrence of disease through reinfection. A similar outcome could be created by the use of anti-inflammatory compounds that suppress both innate and adaptive immune responses. Highly tailored anti-inflammatory drugs such as losartan that block type I angiotensin II receptor, thereby inhibiting inflammation without compromising an adaptive immune response, should be considered. Combination of losartan with passive immunotherapy by means of convalescent plasma in symptomatic patients could be a promising strategy for the prevention or treatment of severe clinical symptoms and will allow patients to develop immunity against the virus.

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References

1. Kim, J. M., Y. S. Chung, H. J. Jo, N. J. Lee, M. S. Kim, S. H. Woo, S. Park, J. W. Kim, H. M. Kim, and M. G. Han. 2020. Identification of coronavirus isolated from a patient in Korea with covid-19. *Osong Public Health Res. Perspect.* 11: 3–7.
2. Bai, Y., L. Yao, T. Wei, F. Tian, D. Y. Jin, L. Chen, and M. Wang. 2020. Presumed asymptomatic carrier transmission of COVID-19. *JAMA.* 323: 1406–1407.
3. Guan, W. J., Z. Y. Ni, Y. Hu, W. H. Liang, C. Q. Ou, J. X. He, L. Liu, H. Shan, C. L. Lei, D. S. C. Hui, et al; China Medical Treatment Expert Group for Covid-19. 2020. Clinical characteristics of coronavirus disease 2019 in China. *N. Engl. J. Med.* 382: 1708–1720.
4. Lauer, S. A., K. H. Grantz, Q. Bi, F. K. Jones, Q. Zheng, H. R. Meredith, A. S. Azman, N. G. Reich, and J. Lessler. 2020. The incubation period of coronavirus disease 2019 (COVID-19) from publicly reported confirmed cases: estimation and application. *Ann. Intern. Med.* 172: 577–582.
5. Mizumoto, K., K. Kagaya, A. Zarebski, and G. Chowell. 2020. Estimating the asymptomatic proportion of coronavirus disease 2019 (COVID-19) cases on board the Diamond Princess cruise ship, Yokohama, Japan, 2020. *Euro Surveill.* DOI: 10.2807/1560-7917.ES.2020.25.10.2000180.
6. Zou, L., F. Ruan, M. Huang, L. Liang, H. Huang, Z. Hong, J. Yu, M. Kang, Y. Song, J. Xia, et al. 2020. SARS-CoV-2 viral load in upper respiratory specimens of infected patients. *N. Engl. J. Med.* 382: 1177–1179.
7. Wu, Z., and J. M. McGoogan. 2020. Characteristics of and important lessons from the coronavirus disease 2019 (COVID-19) outbreak in China: summary of a report of 72 314 cases from the Chinese center for disease control and prevention. *JAMA.* DOI: 10.1001/jama.2020.2648.
8. Vaninov, N. 2020. In the eye of the COVID-19 cytokine storm. *Nat. Rev. Immunol.* 20: 277.
9. Chousterman, B. G., F. K. Swirski, and G. F. Weber. 2017. Cytokine storm and sepsis disease pathogenesis. *Semin. Immunopathol.* 39: 517–528.
10. Huang, C., Y. Wang, X. Li, L. Ren, J. Zhao, Y. Hu, L. Zhang, G. Fan, J. Xu, X. Gu, et al. 2020. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. [Published erratum appears in 2020 *Lancet* 395: 496.] *Lancet* 395: 497–506.
11. Chen, N., M. Zhou, X. Dong, J. Qu, F. Gong, Y. Han, Y. Qiu, J. Wang, Y. Liu, Y. Wei, et al. 2020. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. *Lancet* 395: 507–513.
12. Angele, M. K., S. Pratschke, W. J. Hubbard, and I. H. Chaudry. 2014. Gender differences in sepsis: cardiovascular and immunological aspects. *Virulence* 5: 12–19.
13. Rathod, K. S., V. Kapil, S. Velmurugan, R. S. Khambata, U. Siddique, S. Khan, S. Van Eijl, L. C. Gee, J. Bansal, K. Pitrola, et al. 2017. Accelerated resolution of inflammation underlies sex differences in inflammatory responses in humans. *J. Clin. Invest.* 127: 169–182.
14. Lefèvre, N., F. Corazza, J. Valsamis, A. Delbaere, V. De Maertelaer, J. Duchateau, and G. Casimir. 2019. The number of X chromosomes influences inflammatory cytokine production following toll-like receptor stimulation. *Front. Immunol.* 10: 1052.
15. Libert, C., L. Dejager, and I. Pinheiro. 2010. The X chromosome in immune functions: when a chromosome makes the difference. *Nat. Rev. Immunol.* 10: 594–604.
16. Spolarics, Z. 2007. The X-files of inflammation: cellular mosaicism of X-linked polymorphic genes and the female advantage in the host response to injury and infection. *Shock* 27: 597–604.
17. Carrel, L., and H. F. Willard. 2005. X-inactivation profile reveals extensive variability in X-linked gene expression in females. *Nature* 434: 400–404.
18. Chitnis, S., J. Monteiro, D. Glass, B. Apatoff, J. Salmon, P. Concannon, and P. K. Gregersen. 2000. The role of X-chromosome inactivation in female predisposition to autoimmunity. *Arthritis Res.* 2: 399–406.
19. Spolarics, Z., G. Peña, Y. Qin, R. J. Donnelly, and D. H. Livingston. 2017. Inherent X-linked genetic variability and cellular mosaicism unique to females contribute to sex-related differences in the innate immune response. *Front. Immunol.* 8: 1455.
20. Conti, P., and A. Younes. 2020. Coronavirus COV-19/SARS-CoV-2 affects women less than men: clinical response to viral infection. *J. Biol. Regul. Homeost. Agents.* DOI: 10.23812/Editorial-Conti-3.
21. Casimir, G. J. A., S. Mulier, L. Hanssens, K. Zylberberg, and J. Duchateau. 2010. Gender differences in inflammatory markers in children. *Shock* 33: 258–262.
22. Casimir, G. J., F. Heldenbergh, L. Hanssens, S. Mulier, C. Heinrichs, N. Lefevre, J. Désir, F. Corazza, and J. Duchateau. 2010. Gender differences and inflammation: an in vitro model of blood cells stimulation in prepubescent children. *J. Inflamm. (Lond.)* 7: 28.
23. Simmons, G., J. D. Reeves, A. J. Rennekamp, S. M. Amberg, A. J. Piefer, and P. Bates. 2004. Characterization of severe acute respiratory syndrome-associated coronavirus (SARS-CoV) spike glycoprotein-mediated viral entry. *Proc. Natl. Acad. Sci. USA* 101: 4240–4245.
24. Wang, M., R. Cao, L. Zhang, X. Yang, J. Liu, M. Xu, Z. Shi, Z. Hu, W. Zhong, and G. Xiao. 2020. Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Res.* 30: 269–271.
25. Ducatelle, R., and J. Hoorens. 1984. Significance of lysosomes in the morphogenesis of coronaviruses. *Arch. Virol.* 79: 1–12.
26. Iwasaki, A., and Y. Yang. 2020. The potential danger of suboptimal antibody responses in COVID-19. *Nat. Rev. Immunol.* DOI: 10.1038/s41577-020-0321-6.
27. Souyris, M., C. Cenac, P. Azar, D. Daviaud, A. Canivet, S. Grunenwald, C. Pienkowski, J. Chaumeil, J. E. Mejía, and J.-C. Guéry. 2018. *TLR7* escapes X chromosome inactivation in immune cells. *Sci. Immunol.* DOI: 10.1126/sciimmunol.aap8855.
28. Torcia, M. G., L. Nencioni, A. M. Clemente, L. Civitelli, I. Celestino, D. Limongi, G. Fadigati, E. Perissi, F. Cozzolino, E. Garaci, and A. T. Palamara. 2012. Sex differences in the response to viral infections: TLR8 and TLR9 ligand stimulation induce higher IL10 production in males. *PLoS One* 7: e39853.
29. Meier, A., J. J. Chang, E. S. Chan, R. B. Pollard, H. K. Sidhu, S. Kulkarni, T. F. Wen, R. J. Lindsay, L. Orellana, D. Mildvan, et al. 2009. Sex differences in the toll-like receptor-mediated response of plasmacytoid dendritic cells to HIV-1. *Nat. Med.* 15: 955–959.
30. Zhu, J., T. Zhang, L. Cao, A. Li, K. Zheng, N. Zhang, B. Su, Z. Chen, N. Chen, H. Wu, and Q. He. 2017. Toll like receptor7 polymorphisms in relation to disease susceptibility and progression in Chinese patients with chronic HBV infection. *Sci. Rep.* 7: 12417.
31. Wang, C.-H., H.-L. Eng, K.-H. Lin, C.-H. Chang, C.-A. Hsieh, Y.-L. Lin, and T.-M. Lin. 2011. TLR7 and TLR8 gene variations and susceptibility to hepatitis C virus infection. *PLoS One* 6: e26235.
32. Wu, X.-L., Z.-Y. Li, X.-Y. Bi, H. Zhao, J.-J. Zhao, J.-G. Zhou, Y. Han, Z. Huang, Y.-F. Zhang, and J.-Q. Cai. 2018. Human leukocyte antigen gene polymorphisms are associated with systemic inflammation in hepatitis B virus-related hepatocellular carcinoma. *Cancer Manag. Res.* 10: 2315–2324.
33. Lin, M., H.-K. Tseng, J. A. Trejaut, H.-L. Lee, J.-H. Loo, C.-C. Chu, P.-J. Chen, Y.-W. Su, K. H. Lim, Z.-U. Tsai, et al. 2003. Association of HLA class I with severe acute respiratory syndrome coronavirus infection. *BMC Med. Genet.* 4: 9.
34. Xiong, P., X. Zeng, M. S. Song, S. W. Jia, M. H. Zhong, L. L. Xiao, W. Lan, C. Cai, X. W. Wu, F. L. Gong, and W. Wang. 2008. Lack of association between HLA-A, -B and -DRB1 alleles and the development of SARS: a cohort of 95 SARS-recovered individuals in a population of Guangdong, southern China. *Int. J. Immunogenet.* 35: 69–74.
35. Prompetchara, E., C. Ketloy, and T. Palaga. 2020. Immune responses in COVID-19 and potential vaccines: lessons learned from SARS and MERS epidemic. *Asian Pac. J. Allergy Immunol.* 38: 1–9.
36. Liang, W., W. Guan, R. Chen, W. Wang, J. Li, K. Xu, C. Li, Q. Ai, W. Lu, H. Liang, et al. 2020. Cancer patients in SARS-CoV-2 infection: a nationwide analysis in China. *Lancet Oncol.* 21: 335–337.
37. Fagiolo, U., A. Cossarizza, E. Scala, E. Fanales-Belasio, C. Ortolani, E. Cozzi, D. Monti, C. Franceschi, and R. Paganelli. 1993. Increased cytokine production in mononuclear cells of healthy elderly people. *Eur. J. Immunol.* 23: 2375–2378.
38. Fernández-Atucha, A., A. Izagirre, A. B. Fraile-Bermúdez, M. Kortajarena, G. Larrinaga, P. Martinez-Lage, E. Echevarría, and J. Gil. 2017. Sex differences in the aging pattern of renin-angiotensin system serum peptidases. *Biol. Sex Differ.* 8: 5.
39. Liu, Y., L. M. Yan, L. Wan, T. X. Xiang, A. Le, J. M. Liu, M. Peiris, L. L. M. Poon, and W. Zhang. 2020. Viral dynamics in mild and severe cases of COVID-19. *Lancet Infect. Dis.* DOI: 10.1016/S1473-3099(20)30232-2.
40. Gao, Y., T. Li, M. Han, X. Li, D. Wu, Y. Xu, Y. Zhu, Y. Liu, X. Wang, and L. Wang. 2020. Diagnostic utility of clinical laboratory data determinations for patients with the severe COVID-19. *J. Med. Virol.* DOI: 10.1002/jmv.25770.
41. Mehta, P., D. F. McAuley, M. Brown, E. Sanchez, R. S. Tattersall, and J. J. Manson; HLH Across Speciality Collaboration, UK. 2020. COVID-19: consider cytokine storm syndromes and immunosuppression. *Lancet* 395: 1033–1034.
42. Nicholls, J. M., L. L. M. Poon, K. C. Lee, W. F. Ng, S. T. Lai, C. Y. Leung, C. M. Chu, P. K. Hui, K. L. Mak, W. Lim, et al. 2003. Lung pathology of fatal severe acute respiratory syndrome. *Lancet* 361: 1773–1778.

43. Mahallawi, W. H., O. F. Khabour, Q. Zhang, H. M. Makhdoum, and B. A. Suliman. 2018. MERS-CoV infection in humans is associated with a pro-inflammatory Th1 and Th17 cytokine profile. *Cytokine* 104: 8–13.
44. Wong, C. K., C. W. K. Lam, A. K. L. Wu, W. K. Ip, N. L. S. Lee, I. H. S. Chan, L. C. W. Lit, D. S. C. Hui, M. H. M. Chan, S. S. C. Chung, and J. J. Y. Sung. 2004. Plasma inflammatory cytokines and chemokines in severe acute respiratory syndrome. *Clin. Exp. Immunol.* 136: 95–103.
45. Pedersen, S. F., and Y.-C. Ho. 2020. SARS-CoV-2: a storm is raging. *J. Clin. Invest.* 130: 2202–2205.
46. Liao, Y.-C., W.-G. Liang, F.-W. Chen, J.-H. Hsu, J.-J. Yang, and M.-S. Chang. 2002. IL-19 induces production of IL-6 and TNF-α and results in cell apoptosis through TNF-α. *J. Immunol.* 169: 4288–4297.
47. Abbas, A. K. 1996. Die and let live: eliminating dangerous lymphocytes. *Cell* 84: 655–657.
48. Xu, H., L. Zhong, J. Deng, J. Peng, H. Dan, X. Zeng, T. Li, and Q. Chen. 2020. High expression of ACE2 receptor of 2019-nCoV on the epithelial cells of oral mucosa. *Int. J. Oral Sci.* DOI: 10.1038/s41368-020-0074-x.
49. Fischer, K., P. Hoffmann, S. Voelkl, N. Meidenbauer, J. Ammer, M. Edinger, E. Gottfried, S. Schwarz, G. Rothe, S. Hoves, et al. 2007. Inhibitory effect of tumor cell-derived lactic acid on human T cells. *Blood* 109: 3812–3819.
50. Li, W., M. J. Moore, N. Vasilieva, J. Sui, S. K. Wong, M. A. Berne, M. Somasundaran, J. L. Sullivan, K. Luzuriaga, T. C. Greenough, et al. 2003. Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. *Nature* 426: 450–454.
51. Zhou, P., X. L. Yang, X. G. Wang, B. Hu, L. Zhang, W. Zhang, H. R. Si, Y. Zhu, B. Li, C. L. Huang, et al. 2020. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature* 579: 270–273.
52. Hoffmann, M., H. Kleine-Weber, S. Schroeder, N. Krüger, T. Herrler, S. Erichsen, T. S. Schiergens, G. Herrler, N.-H. Wu, A. Nitsche, et al. 2020. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell* 181: 271–280.e8.
53. Loganathan, S. K., K. Schleicher, A. Malik, R. Quevedo, E. Langille, K. Teng, R. H. Oh, B. Rathod, R. Tsai, P. Samavarchi-Tehrani, et al. 2020. Rare driver mutations in head and neck squamous cell carcinomas converge on NOTCH signaling. *Science* 367: 1264–1269.
54. Zou, X., K. Chen, J. Zou, P. Han, J. Hao, and Z. Han. 2020. Single-cell RNA-seq data analysis on the receptor ACE2 expression reveals the potential risk of different human organs vulnerable to 2019-nCoV infection. *Front. Med.* DOI: 10.1007/s11684-020-0754-0.
55. Sun, P., X. Lu, C. Xu, Y. Wang, W. Sun, and J. Xi. 2020. CD-sACE2 inclusion compounds: an effective treatment for coronavirus disease 2019 (COVID-19). *J. Med. Virol.* DOI: 10.1002/jmv.25804.
56. Zheng, Y. Y., Y. T. Ma, J. Y. Zhang, and X. Xie. 2020. COVID-19 and the cardiovascular system. *Nat. Rev. Cardiol.* 17: 259–260.
57. Ioannidis, I., F. Ye, B. McNally, M. Willette, and E. Flaño. 2013. Toll-like receptor expression and induction of type I and type III interferons in primary airway epithelial cells. *J. Virol.* 87: 3261–3270.
58. Lafferty, E. I., S. T. Qureshi, and M. Schnare. 2010. The role of toll-like receptors in acute and chronic lung inflammation. *J. Inflamm. (Lond.)* 7: 57.
59. Kuba, K., Y. Imai, and J. M. Penninger. 2006. Angiotensin-converting enzyme 2 in lung diseases. *Curr. Opin. Pharmacol.* 6: 271–276.
60. Abassi, Z. A., K. Skorecki, S. N. Heyman, S. Kinaneh, and Z. Armaly. 2020. Covid-19 infection and mortality: a physiologist's perspective enlightening clinical features and plausible interventional strategies. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 318: L1020–L1022.
61. Marshall, R. P., P. Gohlke, R. C. Chambers, D. C. Howell, S. E. Bottoms, T. Unger, R. J. McAnulty, and G. J. Laurent. 2004. Angiotensin II and the fibroproliferative response to acute lung injury. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 286: L156–L164.
62. Benigni, A., P. Cassis, and G. Remuzzi. 2010. Angiotensin II revisited: new roles in inflammation, immunology and aging. *EMBO Mol. Med.* 2: 247–257.
63. Montezano, A. C., A. Nguyen Dinh Cat, F. J. Rios, and R. M. Touyz. 2014. Angiotensin II and vascular injury. *Curr. Hypertens. Rep.* 16: 431.
64. Mateo, T., Y. N. Abu Nabah, M. Abu Taha, M. Mata, M. Cerdá-Nicolás, A. E. I. Proudfoot, R. A. K. Stahl, A. C. Issekutz, J. Cortijo, E. J. Morcillo, et al. 2006. Angiotensin II-induced mononuclear leukocyte interactions with arteriolar and venular endothelium are mediated by the release of different CC chemokines. *J. Immunol.* 176: 5577–5586.
65. Biancardi, V. C., G. F. Bomfim, W. L. Reis, S. Al-Gassimi, and K. P. Nunes. 2017. The interplay between Angiotensin II, TLR4 and hypertension. *Pharmacol. Res.* 120: 88–96.
66. Channappanavar, R., C. Fett, J. Zhao, D. K. Meyerholz, and S. Perlman. 2014. Virus-specific memory CD8 T cells provide substantial protection from lethal severe acute respiratory syndrome coronavirus infection. *J. Virol.* 88: 11034–11044.
67. Temperton, N. J., P. K. Chan, G. Simmons, M. C. Zambon, R. S. Tedder, Y. Takeuchi, and R. A. Weiss. 2005. Longitudinally profiling neutralizing antibody response to SARS coronavirus with pseudotypes. *Emerg. Infect. Dis.* 11: 411–416.
68. Vincent, M. J., E. Bergeron, S. Benjannet, B. R. Erickson, P. E. Rollin, T. G. Ksiazek, N. G. Seidah, and S. T. Nichol. 2005. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virol. J.* 2: 69.
69. Shojania, K., B. E. Koehler, and T. Elliott. 1999. Hypoglycemia induced by hydroxychloroquine in a type II diabetic treated for polyarthritis. *J. Rheumatol.* 26: 195–196.
70. Mohammad, S., M. E. B. Clowse, A. M. Eudy, and L. G. Criscione-Schreiber. 2018. Examination of hydroxychloroquine use and hemolytic anemia in G6PDH-deficient patients. *Arthritis Care Res. (Hoboken)* 70: 481–485.
71. Lee, J. Y., N. Vinayagamoorthy, K. Han, S. K. Kwok, J. H. Ju, K. S. Park, S.-H. Jung, S.-W. Park, Y.-J. Chung, and S.-H. Park. 2016. Association of polymorphisms of cytochrome P450 2D6 with blood hydroxychloroquine levels in patients with systemic lupus erythematosus. *Arthritis Rheumatol.* 68: 184–190.
72. Neafsey, P., G. Ginsberg, D. Hattis, and B. Sonawane. 2009. Genetic polymorphism in cytochrome P450 2D6 (CYP2D6): population distribution of CYP2D6 activity. *J. Toxicol. Environ. Health B Crit. Rev.* 12: 334–361.
73. Elewa, H., and K. J. Wilby. 2017. A review of pharmacogenetics of antimalarials and associated clinical implications. *Eur. J. Drug Metab. Pharmacokinet.* 42: 745–756.
74. Multicenter Collaboration Group of Department of Science and Technology of Guangdong Province and Health Commission of Guangdong Province for Chloroquine in the Treatment of Novel Coronavirus Pneumonia. 2020. [Expert consensus on chloroquine phosphate for the treatment of novel coronavirus pneumonia]. *Zhonghua Jie He He Hu Xi Za Zhi* 43: 185–188.
75. Gao, J., Z. Tian, and X. Yang. 2020. Breakthrough: chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. *Biosci. Trends* 14: 72–73.
76. Kupferschmidt, K., and J. Cohen. 2020. Race to find COVID-19 treatments accelerates. *Science* 367: 1412–1413.
77. Gautret, P., J.-C. Lagier, P. Parola, V. T. Hoang, L. Meddeb, M. Mailhe, B. Doudier, J. Courjon, V. Giordanengo, V. E. Vieira, et al. 2020. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int. J. Antimicrob. Agents* 105949.
78. Gbinigie, K., and K. Frie. 2020. Should chloroquine and hydroxychloroquine be used to treat COVID-19? A rapid review. *BJGP Open.* DOI: 10.3399/bjgpopen20X101069.
79. Gao, W., Y. Xiong, Q. Li, and H. Yang. 2017. Inhibition of toll-like receptor signaling as a promising therapy for inflammatory diseases: a journey from molecular to nano therapeutics. *Front. Physiol.* 8: 508.
80. Schmidt, R. L. J., S. Jutz, K. Goldhahn, N. Witzeneder, M. C. Gerner, D. Trapin, G. Greiner, G. Hoermann, G. Steiner, W. F. Pickl, et al. 2017. Chloroquine inhibits human $CD4^+$ T-cell activation by AP-1 signaling modulation. *Sci. Rep.* 7: 42191.
81. Wu, Y. F., P. Zhao, X. Luo, J. C. Xu, L. Xue, Q. Zhou, M. Xiong, J. Shen, Y. B. Peng, M. F. Yu, et al. 2017. Chloroquine inhibits Ca^{2+} permeable ion channels-mediated Ca^{2+} signaling in primary B lymphocytes. *Cell Biosci.* 7: 28.
82. Furst, D. E. 1996. Pharmacokinetics of hydroxychloroquine and chloroquine during treatment of rheumatic diseases. *Lupus* 5(Suppl. 1): S11–S15.
83. Inungu, J., K. Iheduru-Anderson, and O. J. Odio. 2019. Recurrent Ebolavirus disease in the democratic republic of Congo: update and challenges. *AIMS Public Health* 6: 502–513.
84. PALM Consortium Study Team. 2019. A randomized, controlled trial of Ebola virus disease therapeutics. *N. Engl. J. Med.* 381: 2293–2303.
85. Grein, J., N. Ohmagari, D. Shin, G. Diaz, E. Asperges, A. Castagna, T. Feldt, G. Green, M. L. Green, F.-X. Lescure, et al. 2020. Compassionate use of remdesivir for patients with severe covid-19. *N. Engl. J. Med.* DOI: 10.1056/NEJMoa2007016.
86. Bhatraju, P. K., B. J. Ghassemieh, M. Nichols, R. Kim, K. R. Jerome, A. K. Nalla, A. L. Greninger, S. Pipavath, M. M. Wurfel, L. Evans, et al. 2020. Covid-19 in critically ill patients in the seattle region - case series. *N. Engl. J. Med.* DOI: 10.1056/NEJMoa2004500.
87. Stockman, L. J., R. Bellamy, and P. Garner. 2006. SARS: systematic review of treatment effects. *PLoS Med.* 3: e343.
88. Arabi, Y. M., Y. Mandourah, F. Al-Hameed, A. A. Sindi, G. A. Almekhlafi, M. A. Hussein, J. Jose, R. Pinto, A. Al-Omari, A. Kharaba, et al; Saudi Critical Care Trial Group. 2018. Corticosteroid therapy for critically ill patients with middle east respiratory syndrome. *Am. J. Respir. Crit. Care Med.* 197: 757–767.
89. Lansbury, L. E., C. Rodrigo, J. Leonardi-Bee, J. Nguyen-Van-Tam, and W. Shen Lim. 2020. Corticosteroids as adjunctive therapy in the treatment of influenza: an updated cochrane systematic review and meta-analysis. *Crit. Care Med.* 48: e98–e106.
90. Zha, L., S. Li, L. Pan, B. Tefsen, Y. Li, N. French, L. Chen, G. Yang, and E. V. Villanueva. 2020. Corticosteroid treatment of patients with coronavirus disease 2019 (COVID-19). *Med. J. Aust.* DOI: 10.5694/mja2.50577.
91. Russell, C. D., J. E. Millar, and J. K. Baillie. 2020. Clinical evidence does not support corticosteroid treatment for 2019-nCoV lung injury. *Lancet* 395: 473–475.
92. Cain, D. W., and J. A. Cidlowski. 2017. Immune regulation by glucocorticoids. *Nat. Rev. Immunol.* 17: 233–247.
93. Amici, C., A. Di Caro, A. Ciucci, L. Chiappa, C. Castilletti, V. Martella, N. Decaro, C. Buonavoglia, M. R. Capobianchi, and M. G. Santoro. 2006. Indomethacin has a potent antiviral activity against SARS coronavirus. *Antivir. Ther. (Lond.)* 11: 1021–1030.
94. Zheng, W., W. Fan, S. Zhang, P. Jiao, Y. Shang, L. Cui, M. Mahesutihan, J. Li, D. Wang, G. F. Gao, et al. 2019. Naproxen exhibits broad anti-influenza virus activity in mice by impeding viral nucleoprotein nuclear export. *Cell Rep.* 27: 1875–1885.e5.
95. Kickbusch, I., and G. Leung. 2020. Response to the emerging novel coronavirus outbreak. *BMJ* 368: m406.
96. Bancos, S., M. P. Bernard, D. J. Topham, and R. P. Phipps. 2009. Ibuprofen and other widely used non-steroidal anti-inflammatory drugs inhibit antibody production in human cells. *Cell. Immunol.* 258: 18–28.
97. Kim, H.-J., Y.-H. Lee, S.-A. Im, K. Kim, and C.-K. Lee. 2010. Cyclooxygenase inhibitors, aspirin and ibuprofen, inhibit MHC-restricted antigen presentation in dendritic cells. *Immune Netw.* 10: 92–98.
98. Voiriot, G., Q. Philippot, A. Elabbadi, C. Elbim, M. Chalumeau, and M. Fartoukh. 2019. Risks related to the use of non-steroidal anti-inflammatory

drugs in community-acquired pneumonia in adult and pediatric patients. *J. Clin. Med.* DOI: 10.3390/jcm8060786.
99. Wu, Y. 2020. Compensation of ACE2 function for possible clinical management of 2019-nCoV-induced acute lung injury. *Virol. Sin.* DOI: 10.1007/s12250-020-00205-6.
100. Imai, Y., K. Kuba, S. Rao, Y. Huan, F. Guo, B. Guan, P. Yang, R. Sarao, T. Wada, H. Leong-Poi, et al. 2005. Angiotensin-converting enzyme 2 protects from severe acute lung failure. *Nature* 436: 112–116.
101. Carvalho, T. 2020. COVID-19 research in brief: 28 March to 3 April, 2020. *Nat. Med.* DOI: 10.1038/d41591-020-00008-y.
102. Fedson, D. S. 2016. Treating the host response to emerging virus diseases: lessons learned from sepsis, pneumonia, influenza and Ebola. *Ann. Transl. Med.* 4: 421.
103. Yuan, S. 2015. Statins may decrease the fatality rate of middle east respiratory syndrome infection. *MBio* 6: e01120.
104. Kuba, K., Y. Imai, S. Rao, H. Gao, F. Guo, B. Guan, Y. Huan, P. Yang, Y. Zhang, W. Deng, et al. 2005. A crucial role of angiotensin converting enzyme 2 (ACE2) in SARS coronavirus-induced lung injury. *Nat. Med.* 11: 875–879.
105. Cheng, Y., R. Wong, Y. O. Y. Soo, W. S. Wong, C. K. Lee, M. H. L. Ng, P. Chan, K. C. Wong, C. B. Leung, and G. Cheng. 2005. Use of convalescent plasma therapy in SARS patients in Hong Kong. *Eur. J. Clin. Microbiol. Infect. Dis.* 24: 44–46.
106. Ko, J.-H., H. Seok, S. Y. Cho, Y. E. Ha, J. Y. Baek, S. H. Kim, Y.-J. Kim, J. K. Park, C. R. Chung, E.-S. Kang, et al. 2018. Challenges of convalescent plasma infusion therapy in Middle East respiratory coronavirus infection: a single centre experience. *Antivir. Ther. (Lond.)* 23: 617–622.
107. Hung, I. F., K. K. To, C.-K. Lee, K.-L. Lee, K. Chan, W.-W. Yan, R. Liu, C.-L. Watt, W.-M. Chan, K.-Y. Lai, et al. 2011. Convalescent plasma treatment reduced mortality in patients with severe pandemic influenza A (H1N1) 2009 virus infection. *Clin. Infect. Dis.* 52: 447–456.
108. Ebola-Tx Consortium. 2016. Evaluation of convalescent plasma for Ebola virus disease in Guinea. *N. Engl. J. Med.* 374: 33–42.
109. Duan, K., B. Liu, C. Li, H. Zhang, T. Yu, J. Qu, M. Zhou, L. Chen, S. Meng, Y. Hu, et al. 2020. Effectiveness of convalescent plasma therapy in severe COVID-19 patients. *Proc. Natl. Acad. Sci. USA* 117: 9490–9496.
110. Zhang, B., S. Liu, T. Tan, W. Huang, Y. Dong, L. Chen, Q. Chen, L. Zhang, Q. Zhong, X. Zhang, et al. 2020. Treatment with convalescent plasma for critically ill patients with SARS-CoV-2 infection. *Chest.* DOI: 10.1016/j.chest.2020.03.039.
111. Shen, C., Z. Wang, F. Zhao, Y. Yang, J. Li, J. Yuan, F. Wang, D. Li, M. Yang, L. Xing, et al. 2020. Treatment of 5 critically ill patients with COVID-19 with convalescent plasma. *JAMA.* DOI: 10.1001/jama.2020.4783.
112. Tanne, J. H. 2020. Covid-19: FDA approves use of convalescent plasma to treat critically ill patients. *BMJ* 368: m1256.