TITLE: Is ACE2 a friend or foe in the fight against the COVID19 pandemic?

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ABSTRACT

Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) is responsible for the unprecedented global COVID-19 pandemic that has prompted attempts to mitigate its rapid transmission. A major barrier has been a limited understanding of the mechanisms that underlie its pathogenesis. Angiotensin converting enzyme (ACE) 2 may be an important receptor that the virus targets to establish infection, and this is upregulated with ACE inhibitor and angiotensin-receptor blocker (ARB) therapy. It has been suggested that avoidance of ACE inhibitor/ARB therapy may be advisable due to the theoretical risk of SARS-CoV-2 infection in these patients. A review of the literature to further investigate this possibility corroborated the significance of ACE2 for viral entry of SARS-CoV-2. However, the development of potentially fatal respiratory complications (i.e. acute respiratory distress syndrome) appears to be influenced by a downregulation of ACE2 activity, rather than an increase. The literature suggests ACE2 has a protective role against lung injury by cleaving angiotensin II (Ang II). The effects of this are twofold: accumulated Ang II is associated with more pronounced lung deterioration, and the loss of its degradation products that have anti-inflammatory properties enables unopposed lung destruction to take place. There is emerging evidence that this is also true for SARS-CoV-2 infection; increased viral load is associated with higher Ang II and extent of lung damage. Overall, the evidence indicates that the respiratory complications in severe cases SARS-CoV-2 infection may be due to decreased, rather than increased ACE2 activity. As such, discontinuation of ACE inhibitor/ARB therapy is currently not warranted.

INTRODUCTION

A novel zoonotic strain of coronavirus had emerged in Wuhan, China in December of 2019 that had been responsible for a surge of cases of pneumonia in the region.¹ It had originally been termed the 2019 novel coronavirus (2019-nCoV), and was then renamed as severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2).² The situation rapidly evolved with an unprecedented increase in the number of cases worldwide. This prompted a declaration from the World Health Organization on 30th January 2020, classifying the outbreak as a Public Health Emergency of International Concern.³ One of the major complications of infection is extensive alveolar damage progressing to acute respiratory distress syndrome (ARDS).^{4,5}

The sudden upheaval caused by the SARS-CoV-2 pandemic has precipitated robust attempts to mitigate its rapid transmission. However, the current scarcity of evidence for the pathogenesis of the virus has enabled certain speculative claims to gain traction. A prominent example of this is a recent correspondence published in the *Lancet*, which identified that three separate studies demonstrated that among the most frequent comorbidities in patients requiring hospitalization for SARS-CoV-2 were cardiovascular disease and diabetes mellitus.⁶ The authors noted that these conditions are both frequently treated with angiotensin-converting enzyme (ACE) inhibitor or angiotensin-receptor blocker (ARB) therapy, which may be implicated in the severity of their disease course. They suggest that blockade of ACE1 enzymatic activity from these treatments leads to an increase in expression of ACE2, which serves as the receptor the virus uses to gain entry into cells. Therefore, by increasing the availability of ACE2, these drugs were proposed to facilitate the pathogenesis of SARS-CoV-2. The recommendation was to avoid ACE inhibitors or ARBs in patients with cardiac diseases, hypertension, or diabetes. This article will examine the current literature on the possible role of ACE2 in contributing to the virulence

of SARS-CoV-2, including its involvement in lethal complications such as ARDS, and evaluate the validity of clinical recommendations based on these mechanistic insights.

SIGNIFICANCE OF ACE2 IN SARS-COV-2 PATHOGENESIS

It has previously been established that ACE2 is a key receptor that the closely related SARS-CoV uses to gain intracellular access.⁷ Zhou et al. demonstrated that exposing HeLa cells that either express or do not express ACE2 to SARS-CoV-2 results in viral entry in the ACE2-expressing cells, but not in cells lacking ACE2 expression.⁸ This indicates that ACE2 likely mediates intracellular entry of SARS-CoV-2. The anti-malarial compound chloroquine had previously been shown to be effective in limiting SARS-CoV infectivity in vitro by structurally altering ACE2.⁹ The authors suggested that by reducing the terminal glycosylation of ACE2, chloroquine weakened viral attachment to the cellular surface and prohibited entry. There is recent evidence that chloroquine similarly interferes with the entry step for SARS-CoV-2, and therefore it initially attracted research interest as a potential therapy to combat the current pandemic.¹⁰

LINK BETWEEN ACE2, SARS-COV-2 INFECTIVITY, AND RESPIRATORY COMPLICATIONS

In addition to the role of ACE2 in cellular infiltration by SARS-CoV-2, ACE2 may also halt progression to ARDS, which is one of the known severe complications of SARS-CoV-2 infection. It has been shown that eliminating ACE2 expression in septic mice results in pulmonary damage, evident from increased vascular permeability, edema, and functional decline in the lungs.¹¹ The rationale is that ACE2 normally prevents lung injury by degrading angiotensin II (Ang II), a compound which may trigger inflammation and degradation of the lung

parenchyma through downstream mechanisms (Fig. 1). Ang II has previously been identified as a potential mediator of lung injury by promoting fibroblast proliferation; the structurally altered lung parenchyma in mice with elevated Ang II expression is more vulnerable to damage after exposure to inflammatory mediators.¹² This inflammatory insult also provokes a drastic reduction in lung compliance.

*Receptor blockade of Ang II causes a compensatory increase in its circulating levels

Fig. 1. Overview of ACE inhibitor and ARB effects on Ang II regulation. ACE1 increases circulating Ang II levels, which promotes lung injury. ACE2 promotes Ang II degradation, attenuating lung injury by decreasing Ang II and increasing its anti-inflammatory degradation products. ACE inhibitor therapy simultaneously reduces ACE1 and increases ACE2 activity. ARB therapy increases ACE2 activity, and inhibits Ang II activity via receptor blockade, which increases circulating Ang II levels by reducing negative feedback.¹³

RELEVANCE OF PROTECTIVE EFFECTS OF ACE2 IN HUMANS

The detrimental effects of decreased ACE2 activity and elevated Ang II on respiratory function appears to be evident in humans, as well. Xiaopeng et al. showed ACE2 expression was markedly reduced in lung biopsy specimens obtained from patients with extensive pulmonary fibrosis.¹⁴ Another study revealed that patients with pulmonary fibrosis treated with ACE inhibitor, but not ARB therapy, had slower disease progression.¹⁵ This may be due to lower Ang II with ACE inhibitor therapy due to upstream blockade, and conversely upregulated Ang II levels with ARB therapy from downstream action. The implications of this role in lung damage for viral pneumonia and ARDS is also apparent. Higher mortality rates in influenza A (H7N9) pneumonia was associated with elevated Ang II levels.¹⁶ Death in patients from ARDS is associated with elevated circulating Ang II, and reduced conversion of Ang II to anti-inflammatory peptides by ACE2.¹⁷ Therefore, in addition to buildup of Ang II itself, reduced production of its anti-inflammatory degradation products may also contribute to ARDS. Finally, in a study of 12 SARS-CoV-2 infected patients, half of which developed ARDS, circulating Ang II levels were notably elevated, and linearly correlated with both viral load (and therefore extent of viral infection) and lung injury.¹⁸

CONCLUSION

In summary, ACE2 appears to be a crucial receptor for the infectivity of SARS-CoV-2. Furthermore, ACE2 may have an important role in downregulating Ang II to protect against lung injury. This could at least partly explain the apparent link between decreased ACE2 activity and conditions associated with lung deterioration, such as ARDS and pulmonary fibrosis.

The statement that increased ACE2 promotes transmission of SARS-CoV-2 is likely an oversimplification. In fact, the evidence suggests it is actually decreased ACE2 activity that may

increase susceptibility to severe respiratory complications. Therefore, an alternative explanation may be that viral attachment to ACE2 hinders its ability to cleave Ang II. The result is an accumulation of Ang II that exerts lung damage itself, and the loss of its breakdown products that normally counteract lung inflammation.

The observation that cardiovascular disease and diabetes are frequent comorbidities in severe COVID-19 cases may not be related to ACE inhibitor therapy as originally proposed, but rather the presence of elevated Ang II and decreased ACE2 activity as a common factor for these diseases. Increased ACE2 expression has reduced the progression of diabetes and cardiac disease in mouse models.^{19,20} This provides a potential explanation for the benefit of ACE inhibitor and ARB therapy in these conditions, and that may extend to SARS-CoV-2 infection, in contrast to previous suggestions. It also raises an interesting possibility that the anti-malarial chloroquine, previously identified as having in vitro anti-viral activity against SARS-CoV-2, acts partly through the same mechanism. Increased Ang II has been implicated in the pathogenesis of malaria, and ACE2-mediated conversion of Ang II to certain peptides may protect against parasitic invasion.²¹ The possibility that chloroquine exerts both its anti-malarial and in vitro anti-viral effects in this fashion should be explored further. However, an important caveat is the clinical efficacy of the derivative compound hydroxychloroquine has been questionable. A large observational analysis showed no impact of hydroxychloroquine administration on the rate of intubation or death in patients hospitalized for SARS-CoV-2 infection.²² Furthermore, a randomized placebo-controlled trial demonstrated that hydroxychloroquine use following exposure to SARS-CoV-2 did not prevent symptomatic infection.²³ It is imperative that future studies further examine the involvement of ACE2 and Ang II in a way that will facilitate the development of novel therapeutic options.

REFERENCES

1. Wang D, Hu B, Hu C, et al. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus–infected pneumonia in Wuhan, China. *JAMA*. 2020 Mar;323(11):1061. <https://doi.org/10.1001/jama.2020.1585>
2. Vetter P, Eckerle I, Kaiser L. Covid-19: a puzzle with many missing pieces. *BMJ*. 2020 Feb 19;m627. <https://doi.org/10.1136/bmj.m627>
3. Sohrabi C, Alsafi Z, O'Neill N, et al. World Health Organization declares global emergency: a review of the 2019 novel coronavirus (COVID-19). *Int J Surg*. 2020 Apr;76:71–6. <https://doi.org/10.1016/j.ijsu.2020.02.034>
4. Xu Z, Shi L, Wang Y, Zhang J, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. *Lancet Respir Med*. 2020 Apr;8(4):420–2. [https://doi.org/10.1016/S2213-2600\(20\)30076-X](https://doi.org/10.1016/S2213-2600(20)30076-X)
5. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet*. 2020 Feb;395(10223):497–506. [https://doi.org/10.1016/S0140-6736\(20\)30183-5](https://doi.org/10.1016/S0140-6736(20)30183-5)
6. Fang L, Karakiulakis G, Roth M. Are patients with hypertension and diabetes mellitus at increased risk for COVID-19 infection? *Lancet Respir Med*. 2020 Apr;8(4):e21. [https://doi.org/10.1016/S2213-2600\(20\)30116-8](https://doi.org/10.1016/S2213-2600(20)30116-8)
7. Li W, Moore MJ, Vasilieva N, et al. Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. *Nature*. 2003 Nov;426(6965):450–4. <https://doi.org/10.1038/nature02145>
8. Zhou P, Yang X-L, Wang X-G, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*. 2020 Mar;579(7798):270–3. <https://doi.org/10.1038/s41586-020-2012-7>

9. Vincent MJ, Bergeron E, Benjannet S, et al. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virol J.* 2005;2(1):69. <https://doi.org/10.1186/1743-422X-2-69>
10. Liu J, Cao R, Xu M, et al. Hydroxychloroquine, a less toxic derivative of chloroquine, is effective in inhibiting SARS-CoV-2 infection in vitro. *Cell Discov.* 2020 Mar;6(1):16. <https://doi.org/10.1038/s41421-020-0156-0>
11. Imai Y, Kuba K, Rao S, et al. Angiotensin-converting enzyme 2 protects from severe acute lung failure. *Nature.* 2005 Jul;436(7047):112–6. <https://doi.org/10.1038/nature03712>
12. Wang J, Chen L, Chen B, et al. Chronic Activation of the Renin-Angiotensin System Induces Lung Fibrosis. *Sci Rep.* 2015 Dec;5(1):15561. <https://doi.org/10.1038/srep15561>
13. Agata J, Ura N, Yoshida H, et al. Olmesartan Is an Angiotensin II Receptor Blocker with an Inhibitory Effect on Angiotensin-Converting Enzyme. *Hypertens Res.* 2006 Nov;29(11): 865–74. <https://doi.org/10.1291/hypres.29.865>
14. Li X, Molina-Molina M, Abdul-Hafez A, et al. Angiotensin converting enzyme-2 is protective but downregulated in human and experimental lung fibrosis. *Am J Physiol Lung Cell Mol Physiol.* 2008 Jul;295(1):L178–85. <https://doi.org/10.1152/ajplung.00009.2008>
15. Kreuter M, Lederer DJ, Molina-Molina M, et al. Association of Angiotensin Modulators With the Course of Idiopathic Pulmonary Fibrosis. *Chest.* 2019 Oct;156(4):706–14. <https://doi.org/10.1016/j.chest.2019.04.015>
16. Huang F, Guo J, Zou Z, et al. Angiotensin II plasma levels are linked to disease severity and predict fatal outcomes in H7N9-infected patients. *Nat Commun.* 2014 May;5(1):3595. <https://doi.org/10.1038/ncomms4595>

17. Reddy R, Asante I, Liu S, et al. Circulating angiotensin peptides levels in Acute Respiratory Distress Syndrome correlate with clinical outcomes: A pilot study. PLoS ONE. 2019 Mar;14(3):e0213096. <https://doi.org/10.1371/journal.pone.0213096>
18. Liu Y, Yang Y, Zhang C, et al. Clinical and biochemical indexes from 2019-nCoV infected patients linked to viral loads and lung injury. Sci China Life Sci. 2020 Feb;63(3):364–74. <https://doi.org/10.1007/s11427-020-1643-8>
19. Bindom SM, Hans CP, Xia H, et al. Angiotensin I-Converting Enzyme Type 2 (ACE2) Gene Therapy Improves Glycemic Control in Diabetic Mice. Diabetes. 2010 Oct;59(10):2540–8. <https://doi.org/10.2337/db09-0782>
20. Huentelman MJ, Grobe JL, Vazquez J, et al. Protection from angiotensin II-induced cardiac hypertrophy and fibrosis by systemic lentiviral delivery of ACE2 in rats: ACE2 prevents cardiac hypertrophy and fibrosis. Exp Physiol. 2005 Sep;90(5):783–90. <https://doi.org/10.1113/expphysiol.2005.031096>
21. Silva LS, Silva-Filho JL, Caruso-Neves C, et al. New Concepts in Malaria Pathogenesis: The Role of the Renin-Angiotensin System. Front Cell Infect Microbiol. 2016 Jan;5(1):103 <https://doi.org/10.3389/fcimb.2015.00103>
22. Geleris J, Sun Y, Platt J, et al. Observational study of hydroxychloroquine in hospitalized patients with Covid-19. N Engl J Med. 2020 Jun; 382(25):2411–8. <https://doi.org/10.1056/NEJMoa2012410>
23. Boulware DR, Pullen MF, Bangdiwala AS, et al. A Randomized Trial of Hydroxychloroquine as Postexposure Prophylaxis for Covid-19. N Engl J Med. Epub 2020 Jun. <https://doi.org/10.1056/NEJMoa2016638>