Risks of ACE Inhibitor and ARB Usage in COVID-19: Evaluating the Evidence

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Concerns have been raised regarding the safety of angiotensin converting enzyme inhibitors (ACEIs) and angiotensin receptor blockers (ARBs) in patients with coronavirus disease of 2019 (COVID-19), based on the hypothesis that such medications may raise expression of ACE2, the receptor for severe acute respiratory syndrome-coronavirus 2 (SARS-CoV-2). We conducted a literature review of studies (*n* = 12) in experimental animals and human subjects (*n* = 12) and evaluated the evidence regarding the impact of administration of ACEIs and ARBs on ACE2 expression. We prioritized studies that assessed ACE2 protein expression data, measured directly or inferred from ACE2 activity assays. The findings in animals are inconsistent with respect to an increase in ACE2 expression in response to treatment with ACEIs or ARBs. Control/sham animals show little to no effect in the plurality of studies. Those studies that report increases in ACE2 expression tend to involve acute injury models and/or higher doses of ACEIs or ARBs than are typically administered to patients. Data from human studies overwhelmingly imply that administration of ACEIs/ARBs does not increase ACE2 expression. Available evidence, in particular, data from human studies, does not support the hypothesis that ACEI/ARB use increases ACE2 expression and the risk of complications from COVID-19. We conclude that patients being treated with ACEIs and ARBs should continue their use for approved indications.

There has been much recent debate regarding the use of angiotensin converting enzyme inhibitors (ACEIs) and angiotensin receptor blockers (ARBs) in patients with coronavirus disease of 2019 (COVID-19),[1–7] thus prompting concern among patients and health care providers. The basis of this concern involves whether ACEIs/ARBs increase expression of ACE2, the primary cellular receptor for the severe acute respiratory syndrome-coronavirus 2 (SARS-CoV-2), thereby possibly increasing severity of the infection. Published correspondences/letters, such as those cited above, typically make assertions on a relationship between ACEI/ARB use and ACE2 expression levels but lack a detailed, comprehensive breakdown of available data in humans and animals. The strength of the experimental data is, thus, unclear regarding ACEI/ARB use and ACE2 protein expression.

To address this issue, we have reviewed key literature by assessing studies conducted in experimental animals (primarily rats) and humans, in order to obtain a more comprehensive picture of what the available data convey. We focus our discussion on studies for which ACE2 protein data are available (either via direct measurement or inferred from ACE2 activity assays), as tissue ACE2 mRNA expression seems to only weakly correlate with protein expression, as shown from data in the Human Protein Atlas[8] (proteinatlas.org), results from human renal samples[9] and studies in experimental animals (discussed below and in refs. 10,11).

We identified relevant studies by searches in PubMed and Google Scholar for all available literature. Combinations of search terms were used to maximize the identification of relevant studies. Search terms included "ACE inhibitor," "angiotensin receptor blocker," "ACE inhibition," "ACE2," "ACE2 expression," "ACE2 protein expression," "ACE2 activity," "humans," "patients," "lung," "heart," and "kidney." We imposed no limits on when studies were performed. We manually curated each relevant hit to ensure the data were from articles with original research and/or meta-analysis and included quantitative/normalized ACE2 protein expression/activity and involved studies with tissues relevant to COVID-19/SARS-CoV-2 infection.

WHAT DO DATA FROM ANIMAL AND HUMAN STUDIES IMPLY FOR THE RISK OF ACE INHIBITORS AND ARBS IN PATIENTS WITH COVID-19?

We focused our assessment on studies that evaluated ACE2 levels in tissues/cells involved in infection by SARS-CoV-1 or SARS-CoV-2. Data from 12 animal studies are summarized in **Table 1**, which shows the effect of ACEI/ARB use on ACE2 protein expression/activity in animals (primarily rats). We used conversion factors based on the principles of allometric scaling to evaluate doses in animals relative to their use in humans.[12] For studies in which this was relevant, we used the human equivalent dose (assuming a 60 kg human[12]) for doses of ACEI/ARBs. The conversion factor for rats was 6.2 (i.e., doses in mg/kg were divided by 6.2, then multiplied by 60 kg for a human equivalent dose) and 1.1 for pigs. We obtained recommended doses for humans using approved labeling from the US Food and Drug Administration (FDA) website (fda.gov). These recommended human doses are provided in **Table 2**. For most of the drugs, the maximum doses indicated in **Table 2** are generally not administered to humans, such that treatment of animals with equivalent (or larger) doses, (especially in models of acute dosing, as in refs. 13 and 14 discussed

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Table 1 Studies in animals that have assessed ACE2 protein expression in response to ACEI/ARB treatment

Source	Study details	Effect of ACEI/ ARB on ACE2
Ferrario *et al.* [10]	Lewis rats were treated with losartan (an ARB) or lisinopril (an ACEI) 10 mg/kg/day (HED = 96.8 mg/day), for 20 days. ACE2 activity was measured in membranes from the renal cortex.	Lisinopril or losartan treatment were both associated with increases in ACE2 activity but used in combination, did not produce this effect
Ocaranza *et al.*[15]	Sprague Dawley rats were used in a myocardial infarction model, via coronary ligation. The ACEI enalapril (10 mg/kg/day; HED = 96.8 mg/day) was administered for 8 weeks postsurgery. Plasma ACE2 activity was measured.	Enalapril increased plasma ACE2 by ~ 14% and ~ 36% in sham and MI animals, respectively
Hamming *et al.*[16]	Renal ACE2 activity was assayed in Wistar rats, controls or on low sodium diet along with lisinopril (75 g/L) in drinking water for 3 weeks	Renal ACE2 activity was unchanged with ACEI treatment in either group
Velkoska *et al.*[21]	ACE2 activity was assessed in kidneys from Sprague Dawley rats with subtotal nephrectomy and given ramipril (ACEI, 1 mg/kg/day; HED = 9.68 mg/day) for 10 days	ACE2 activity in renal cortex and medulla was unchanged by ACEI treatment in control rats and increased ~ 50% with nephrectomy
Han *et al.*[17]	ACE2 protein expression in lungs was measured in Sprague Dawley rats with cigarette smoke-induced lung damage. Rats were treated with losartan (10 or 30 mg/kg/day; HED = 96.8 or 290 mg/day) for 6 months	ACE2 expression was unchanged in control rats by either dose of losartan. Animals exposed to cigarette smoke had reduced ACE2, which losartan treatment restored.
Wösten-van Asperen *et al.*[13]	Sprague Dawley rats were used in a LPS-induced model of ARDS. Rats were given losartan (2.5 mg/kg/h during 4 hours of ventilation; HED = 96.8 mg). ACE2 protein expression in the lung was measured 24 hours after inducing ARDS with LPS.	Losartan administration decreased ACE2 activity in control animals (unclear if/how statistics were performed). After induction of ARDS, ACE2 levels decreased and were restored to normal by losartan
Burrell *et al.*[18]	ACE2 activity and protein expression were assayed in tissues from SD rats 28 days after subtotal nephrectomy and which received ramipril (an ACEI, 1 mg/kg/day; HED = 9.68 mg/day)	Ramipril had no effect on ACE2 in cardiac or renal (cortex or medullary) tissue. ACE2 activity was reduced by nephrectomy; ramipril restored ACE2 activity to control levels in renal cortex, but not in medullary or cardiac tissue.
Burchill *et al.*[19]	ACE2 protein expression was assessed in cardiac tissue in an MI model in SD rats. Ramipril (1 mg/kg/day; HED = 9.68 mg/day) and valsartan (ARB, 10 mg/kg/day; HED = 96.8 mg/day) were given for 28 days postcoronary artery ligation.	ACE2 expression was not altered but may have decreased in viable myocardium border or infarct zones, (unclear statistical analysis)
Yang *et al.*[11]	Spontaneously hypertensive rats were treated with enalapril (15 mg/kg/day; HED = 145.2 mg/day) for 4 weeks. Cardiac ACE2 protein expression was measured.	ACE2 mRNA expression was increased but ACE2 protein expression did not change with ACEI treatment
Zhang *et al.*[37]	Cardiac ACE2 protein was assayed from SD rats with cardiac remodeling from aortic constriction and treated with losartan (30 mg/kg/day; HED = 290.3 mg/day) or enalapril (20 mg/kg/day; HED = 193.5 mg/day) for 20 days, starting 4 days after surgery	ACE2 cardiac protein expression was increased (~ 3-fold) with both drugs in rats with cardiac remodeling; data were not provided for animals with sham surgery
Li *et al.*[14]	SD rats underwent LPS-stimulated lung injury. Simultaneous with intravenous injection of LPS, animals were administered captopril (50 mg/kg; HED = 483.9 mg). After 8 hours, samples were collected and ACE2 protein expression was measured in the lungs.	ACE2 protein expression was elevated in the lungs, in control rats (~ 35%) and those with LPS-induced lung injury (~ 2-fold)
Wang *et al.*[20]	Pigs were used to study effects of ACEIs on cardiac arrest and resuscitation. Enalapril (0.2 mg/kg; HED = 10.9 mg) was perfused for 30 minutes, followed by surgery. Myocardial ACE2 protein expression was assayed in samples collected 6 hours postsurgery.	Compared with saline-infused controls, enalapril did not increase ACE2 levels; enalapril was not administered to sham rats

ACEI, angiotensin converting enzyme inhibitor; ARB, angiotensin receptor blocker; ARDS, acute respiratory distress syndrome; HED, human equivalent dose; LPS, lipopolysaccharide; MI, myocardial infarction; SD, Sprague Dawley.

in **Table 1**) raise concern about their relevance to ACEI/ARB administration to patients.

In summary, three studies[10,14,15] in animals reported an increase in all treatment groups of ACE2 protein expression/activity with ACEI/ARB treatment but in one such study,[10] combined ARB/ACEI treatment did not show this effect. The other two studies reported relatively small effects (< 40% increase in ACE2 protein expression in control/sham animals). All of these studies used high doses of ACEIs/ARBs, as noted above. By contrast, six studies[11,16–20] found little or no change in ACE2 expression with ACEI/ARB treatment. In three studies,[13,14,21] treatment with ACEI/ARB had no effect or a decrease in ACE2 protein expression/activity in control/sham animals; increased ACE2 expression was only observed following experimental exposure,

such as lung injury, myocardial infarction, etc. In nearly all studies that reported an increase in ACE2, doses of ACEIs/ARBs used were greater than equivalent doses typically administered to patients. We identified only one study[21] in which ACEI/ARB use increased ACE2 protein expression at doses typically used to treat patients, albeit these changes only occurred after exposure to acute injury (subtotal nephrectomy). Overall, the studies with experimental animals do not provide consistent evidence for an effect of ARB/ACEI administration on ACE2 protein expression, especially in contexts that model drug administration in humans.

A challenge with assessing the animal studies in **Table 1** is that, although doses were provided for the drugs, pharmacokinetic studies were not performed to ascertain peak serum concentration (C_{max}) and area under the curve (AUC), which would allow more precision in evaluating the translational relevance of findings. However, data from other studies provide insight into the pharmacokinetics in rats, at doses comparable to those used in the studies in **Table 1**. **Table 2** shows estimates for C_{max} for the drugs used in the studies in **Table 1**, in humans (at therapeutic doses, as indicated) and rats (at doses comparable to those in **Table 1**). The differences in AUC for these drugs between humans and rats mirror differences in C_{max}; we refer readers to the indicated references for further details. With the exception of a study in a nephrectomy model,[21] animal data (including all data for control animals) that show a relationship between ACE2 expression and use of ACEIs/ARBs involved drug treatments associated with plasma concentrations far in excess (often more than one order of magnitude) of values normally seen in patients. Absent exposure-response studies with more clinically appropriate drug concentrations, we conclude that the bulk of the animal data regarding ACE2 expression with ACEI/ARB administration has dubious translational relevance.

Evidence of changes in ACE2 protein in human subjects/patients (**Table 3**) are derived from studies that assessed ACE2 protein concentration or enzymatic activity in urine or serum/plasma. Of the 11 studies summarized in **Table 3**, seven showed no effect of ARB/ACEI use on ACE2 protein levels in any condition/patient grouping. One study[22] documented a small increase in serum ACE2 attributable to use of ACEIs among patients with type-1 diabetes but found no effect from use of ARBs. Another study[23] found a slightly larger proportional decrease in urinary ACE2 in patients with type-2 diabetes using ACEIs/ARBs but did not distinguish between effects from use of ACEIs or ARBs. In one study,[24] the investigators observed that subjects using the ARB olmesartan had increased ACE2 levels, but several other ARBs and ACEIs had no effect. In one study,[25] ACEI use had no effect in control, patients with stage 3–5 chronic kidney disease or those on dialysis; ARB only had a small effect in patients on dialysis. Besides these quantitative data (in **Table 3**), another study used immunohistochemical analysis to assess ACE2 protein expression in the kidneys and found no ACEI-dependent effect.[26] Together, these 12 studies in humans imply a lack of association between ACE2 protein expression and the use of ARBs or ACEIs and support the idea that ACEIs/ARBs are unlikely to raise ACE2 or be harmful in the context of COVID-19 infection.

TESTING THE HYPOTHESIS THAT ACE INHIBITORS AND ARBS ENHANCE THE SEVERITY OF SARS-COV2 INFECTION

What would constitute strong, supportive evidence for the hypothesis that ACEI/ARB usage is a risk factor in the setting of SARS-CoV2 infection? Findings to help support that hypothesis would include: (i) replication of a prominent effect in multiple animal studies and models; (ii) evidence that tissues with low expression of ACE2 have prominent increases in its expression and activity following ACEI/ARB treatment; (iii) data documenting that increases in ACE2 expression in response to ACEI/ARB treatment enhance the ability of the SARS-CoV-2 virus to infect cells; (iv) findings from human studies of statistically significant relationships between ACEI/ARB usage and ACE2 expression/activity; and (v) epidemiological data showing that patients with COVID-19 administered ACEIs/ARBs have increased morbidity and mortality, ideally with a dose-response relationship for such outcomes.

Table 2 FDA-recommended doses of the ACEIs/ARBs discussed in Table 1, along with pharmacokinetics data in humans and rats

Drug	Typical initial daily adult dose	Maximum daily adult dose	C_{max} and AUC in humans	C_{max} and AUC in rats
Losartan	50 mg	100 mg	250 ng/mL C_{max}, 1,000 ng.hour/mL $AUC_{0-\infty}$ for 50 mg dose[38]	1,260 ng/mL C_{max}, 8250 ng.hour/mL AUC_{0-24h} for 10 mg/kg dose; HED = 96.8 mg[39]
Enalapril	5 mg	40 mg	62 ng/mL C_{max}, 107.5 ng.hour/mL $AUC_{0-\infty}$ for 10 mg dose[40]	1329 ng/mL C_{max}, 7458 ng.hour /mL $AUC_{0-\infty}$ for 15 mg/kg dose; HED = 145.2 mg[41]
Lisinopril	10 mg	40 mg	38 ng/mL C_{max}, 1,000 ng.hour/mL $AUC_{0-\infty}$ for 10 mg dose[42]	6,400 ng/mL C_{max}, 9,800 ng.hour/mL $AUC_{0-\infty}$ for 5 mg/kg dose; HED = 48.4 mg[43]
Ramipril	2.5 mg	20 mg	52.2 ng/mL C_{max}, 104 ng.hour/mL $AUC_{0-\infty}$ at 10 mg dose[42]	50.45 ng/mL C_{max}, 125 ng.hour/mL $AUC_{0-\infty}$ for 1 mg/kg dose; HED = 9.68 mg[44]
Captopril	50 mg	450 mg	800 ng/mL C_{max} for 100 mg dose[45] (AUC unavailable in ref. 45)	5,000 ng/mL for 30 mg/kg dose; HED = 290.4 mg[45] (AUC unavailable in ref. 45)
Valsartan	80 mg	320 mg	2,000 ng/mL C_{max}, 6,480 ng.hour/mL $AUC_{0-\infty}$ for 80 mg dose[38]	~ 3,700 ng/mL C_{max}, ~ 19,500 ng.hour/mL AUC_{0-24} for 4.71 mg/kg dose; HED = 45.6 mg[46]

ACEI, angiotensin converting enzyme inhibitor; ARB, angiotensin receptor blocker; $AUC_{0-\infty}$, area under the concentration-time curve from zero to infinity; AUC_{0-24h}, 0–24-hour area under the concentration-time curve; C_{max}, peak serum concentrations; HED, human equivalent dose.

Table 3 Studies in humans of the relationship between ACEI/ARB use and ACE2 protein expression

Source	Details of study	Effect of ACEI/ ARB on ACE2
Mizuiri *et al.*[47]	Urinary ACE2 protein levels were measured in 190 patients with chronic kidney disease and 36 healthy subjects	No significant difference in urinary ACE2 was observed in response to treatment with ACEI and ARB
Furuhashi *et al.*[24]	Urinary ACE2 protein concentration was assayed in 617 subjects, including 101 subjects who did not use any medication and 100 hypertensives treated with various drugs	Enalapril, losartan, valsartan, candesartan, valsartan, and telmisartan had no effect. Olmesartan increased urinary ACE2.
Liang *et al.*[23]	Urinary ACE2 protein concentration was assessed in 132 patients with type-2 diabetes and 34 healthy volunteers	Patients with hypertension had an ~ 40% decrease in urinary ACE2 if treated with inhibitors of renin-angiotensin signaling, compared with hypertensive patients not taking such medications
Mariana *et al.*[48]	Urinary ACE2 protein levels were measured via ELISA in 75 patients with type-2 diabetes	Use of ARBs or ACEIs had no effect on urinary ACE2 levels
Epelman *et al.*[49]	Plasma ACE2 activity was assayed from 228 patients with heart failure	No association was found between ACEI/ARB use and ACE2 levels
Soro-Paavonen *et al.*[22]	Serum ACE2 activity was measured in 859 patients with type-1 diabetes and 99 healthy control subjects	ACE2 was increased ~ 10 to 20% (higher in women) in patients with diabetes using ACEIs. No association was found between ARB usage and ACE2 levels.
Ortiz-Perez *et al.*[50]	Serum ACE2 activity was assayed in 95 patients with ST-elevation myocardial infarction and 22 control subjects	No association was found between ACEI use and ACE2 levels. ARB usage was not discussed.
Anguiano *et al.*[25]	Plasma ACE2 activity was measured in n = 568 control subjects, n = 1458 with stage 3–5 chronic kidney disease, and n = 546 patients on dialysis. Multivariate regression analysis was performed to identify which factors influenced ACE2.	ACEI use had no effect on ACE2 in any group. ARB use did not predict ACE2 activity in control or stage 3–5 patients; in patients on dialysis ARB use had a small effect raising ACE2 activity.
Uri *et al.*[51]	Serum ACE2 activity was assayed in 141 healthy subjects, 239 hypertensive patients, and 188 patients with heart failure of different types	Logistic regression analysis showed that ACEI and ARB usage had no association with ACE2 levels
Walters *et al.*[52]	Plasma ACE2 activity was assessed in 25 control subjects and 88 patients with atrial fibrillation	No association was found between ACE2 levels and ACEI/ARB use
Ramchand *et al.*[53]	Plasma ACE2 activity was measured in 79 patients with obstructive coronary artery disease	ACE2 levels had no association with use of ACEIs or ARBs

Entries are ordered chronologically, first for studies in urine and then for studies in circulating ACE2.
ACEI, angiotensin converting enzyme inhibitor; ARB, angiotensin receptor blocker; ELISA, enzyme-linked immunosorbent assay.

How well do the available data provide such evidence? (i) The hypothesis that ACE2 expression increases with ACEI/ARB use is not supported by the plurality of available data from animal studies. (ii) There is no available evidence for *de novo* expression of ACE2 expression in response to ACEIs/ARBs in tissues with low expression. (iii) The affinity of SARS-CoV-2 for ACE2 is very high, approximately fourfold greater than SARS-CoV-1[27] or higher; some studies suggest an order-of-magnitude higher affinity for SARS-CoV-2.[28] It is unclear if small or modest perturbations in ACE2 expression impact the infectivity of SARS-COV-2. Moreover, ACE2 levels may decrease with age[29,30] and diabetes,[31] yet elderly/diabetic subjects are more vulnerable than younger individuals to COVID-19.[32] Modest changes in ACE2 expression (< 2-fold, from most preclinical data discussed above) may not meaningfully impact on the high infectivity of SARS-CoV-2 in host tissues. (iv) Data from human studies suggest that treatment with ACEIs/ARBs produces little or no effect on urinary or circulating ACE2 levels. As a caveat, changes in ACE2 levels in serum or urine may not reflect changes in tissue. (v) Epidemiological studies are slowly becoming available in matched groups of patients infected with SARS-CoV-2 who have or have not been taking ACEIs/ARBs. One preliminary study[33] examined 28 patients with severe COVID-19 and 18 patients with mild disease, all of whom also had hypertension. ARB use was associated with a reduction in risk of severe COVID-19 disease, morbidity, and mortality, a result that contradicts the hypothesis that ARB/ACEI use is harmful. However, only small numbers of patients were assessed. A larger retrospective study from 1,128 patients with COVID-19 in China with hypertension found a > 2-fold reduction in mortality in patients administered ACEIs/ARBs compared with patients not receiving those drugs.[34] An additional, smaller retrospective study also noted that hypertensive patients with COVID-19 receiving ACEI/ARB treatment had a lower rate of severe COVID-19 disease, a trend toward reduced blood levels of interleukin-6 plus reduced viral load and increased counts of cytotoxic T cells.[35] None of these three retrospective studies found an elevation of risk with ACEI/ARB use.

Based on the data summarized above, we conclude that current evidence, especially from human studies (**Table 3**), does not support the idea that treatment with ACEIs or ARBS produces

pathophysiologically relevant increases in ACE2 protein abundance. The hypothesis that the use of these drugs increases SARS-CoV-2 virus infectivity and/or severity of COVID-19 is, therefore, not supported by the available evidence. It would thus seem prudent for patients to continue receiving these medications, as recently recommended by multiple health associations[2] and other publications.[3,6]

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CONFLICTS OF INTEREST
The authors declared no conflict of interest.

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