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**New perspectives on angiotensin-converting enzyme 2 and its related diseases**

Liu LP *et al*. Angiotensin-converting enzyme 2

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**Abstract**

Since the worldwide outbreak of coronavirus disease 2019, angiotensin-converting enzyme 2 (ACE2) has received widespread attention as the cell receptor of the severe acute respiratory syndrome coronavirus 2 virus. At the same time, as a key enzyme in the renin-angiotensin-system, ACE2 is considered to be an endogenous negative regulator of vasoconstriction, proliferation, fibrosis, and proinflammation caused by the ACE-angiotensin II-angiotensin type 1 receptor axis. ACE2 is now implicated as being closely connected to diabetes, cardiovascular, kidney, and lung diseases, and so on. This review covers the available information on the host factors regulating ACE2 and discusses its role in a variety of pathophysiological conditions in animal models and humans.

**Key Words:** Angiotensin-converting enzyme 2; COVID-19; Salt; Renin-angiotensin-system inhibitors; Diabetes and cardiovascular disease; Renal and lung disease

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**Core Tip:** Angiotensin-converting enzyme 2 (ACE2) as the key cell receptor for the severe acute respiratory syndrome coronavirus 2 virus has received widespread attention. This paper will review the new perspectives on ACE2, covers available information on the host regulative factors of ACE2, and discusses its role in a variety of pathophysiological conditions. This review will help us with a better understanding of the biological function and role of ACE2 in coronavirus disease 2019 and its treatment.

**INTRODUCTION**

Angiotensin-converting enzyme 2 (ACE2), as the key enzyme in the renin angiotensin system (RAS) and key cell receptor for the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) virus, has received considerable attention[1] since coronavirus disease 2019 (COVID-19) has spread worldwide. Patients with diabetes and renal and cardiovascular diseases are widely treated with ACE inhibitors (ACEIs)/angiotensin II receptor blockers (ARBs), which are deemed to potentially increase ACE2 expression in the body; the consequent increased expression of ACE2 could facilitate infection with SARS-CoV-2. The paradox of the use of ACEI/ARB treatment to interfere with the RAS because ACE2 reduces inflammation (which has been suggested as a therapy for inflammatory lung diseases, diabetes, and hypertension) and the probable contribution of ACE2 to an increased risk of COVID-19 have puzzled clinicians and attracted much attention[2,3]. Therefore, it is necessary to further understand the characteristics of ACE2 and accurate details of the molecular mechanisms of ACE2 that underlie these phenomena. This review will focus on the biological function of ACE2 and its host regulatory factors. Moreover, we will discuss the role of ACE2 in a variety of pathophysiological conditions, including cardiovascular, kidney, and lung diseases and diabetes in animal models and humans. We aim to provide new perspectives on ACE2 and help us better understand the role of ACE2 in SARS-CoV-2 infection and its treatment nowadays (Figure 1).

**biochemical characteristics and distribution of ACE2**

ACE2 is a type I membrane protein that includes an N-terminal peptidase domain (PD) and a C-terminal collectrin-like domain. The PD of ACE2 provides a direct binding site for the S protein of coronavirus[4]. ACE2 was discovered in 2000 when Tipnis *et al*[5] and Donoghue *et al*[6] cloned ACE2 from a human heart failure (HF) cDNA library and a human lymphoma cDNA library and found to be a homolog of the ACE gene. The gene is located in the Xp22 region and has a total length of approximately 39.98 kb, including 18 exons and 17 introns. ACE2 can be hydrolyzed by depolymerase into soluble ACE2, and therefore, ACE2 has two forms, the membrane-linked type and the soluble type. Membrane-linked ACE2 is an extracellular enzyme that is distributed on the cell membrane surface and consists of four parts: The N-terminal signal peptide region, the zinc binding motif (amino acid residues 374 to 378), the transmembrane region (amino acids 740 to 768), and the C-terminal intracellular domain. Dissolved ACE2 is distributed mainly in the plasma and urine due to the lack of a transmembrane region and a C-terminal intracellular domain[5]. ACE2 is widely expressed in many organs, including the heart, kidney, testis, adipose tissue, brain tissue, vascular smooth muscle cells, gastrointestinal tract, and lung (type 2 alveolar epithelial cells) and is highly specific in tissues[7].

**Functions of ACE2**

ACE2 is a key enzyme in the RAS and is considered to be an endogenous negative regulator of vasoconstriction, proliferation, fibrosis, and proinflammation caused by the ACE-angiotensin II (Ang II)-angiotensin type 1 receptor (AT1R) axis[8]. In the RAS, ACE cuts Ang I to convert it into Ang II, and then Ang II binds to the G protein-coupled receptor AT1R, which causes vasoconstriction and increased blood pressure[9]. The function of ACE2 is opposite to that of ACE. ACE2 cuts Ang II and converts it into angiotensin 1-7 (Ang 1-7), which can lower blood pressure[10]. Increasing evidence demonstrates that ACE2 plays an important role in a variety of pathophysiological conditions. In the tumor microenvironment, ACE2 acts on Ang II to produce endogenous Ang 1-7. Ang 1-7 binds to the Mas receptor (MasR) and exerts antitumor effects such as antiproliferation, antiangiogenesis, anti-invasion, and antimetastasis effects through a series of signal transduction pathways. In diabetes, upregulation of ACE2 can improve hyperglycemia, and Ang 1-7 can improve metabolic syndrome through glucose intake and oxidative stress related to insulin resistance[11]. The expression of ACE2 is related to atherosclerosis[12]. The overexpression of ACE2 can improve endothelial-dependent vascular relaxation, increase the proliferative activity of endothelial cells, and facilitate the migration of endothelial cells. ACE2 exerts anti-infective properties through Ang 1-7[13]. The decrease in aortic ACE2 expression increases the expression of proinflammatory factors, such as tumor necrosis factor α, interleukin-6, monocyte chemotactic protein 1, vascular cell adhesion molecule 1, matrix metalloproteinase (MMP)-2 and MMP-9, which are conducive to the adhesion of leukocytes to endothelial cells and blood vessel walls. In addition, ACE2 can also regulate the adhesion of macrophages to endothelial cells[14]. The combination of Ang 1-7 and MasR can also inhibit the formation of thrombi[15] and liver fibrosis[16]. In ischemic stroke, Ang 1-7 can reduce the area of cerebral infarction and brain dysfunction by regulating the release of nitric oxide from different sources to protect the brain. In addition, Ang 1-7 in brain tissue can regulate learning and memory functions. In the reproductive system, Ang 1-7 can regulate endometrial function, spermatogenesis, follicular maturation, ovulation, and pregnancy-related processes[11]. ACE2 can also regulate the immunity of intestinal epithelial cells through amino acid homeostasis, expression of antimicrobial peptides, and regulation of the balance of intestinal microbes[17].

**ACE2 and salt**

It has been widely demonstrated that a high-sodium (HS) diet activates the RAS. Animal experiments have proven that salt intake is a powerful regulator of ACE2 expression in animal models[18]. Studies by Samuel *et al*[19] found that the relative expression patterns of ACE and AT1R increased, renin levels decreased, and ACE2, AT2R, and MasR remained unaltered in HS-fed lean Zucker rats. On the other hand, HS intake caused an increase in the cortical expression of ACE and a decrease in ACE2, accompanied by increased blood pressure. Elevated blood pressure is associated with a significant increase in Ang II levels in the renal cortex in obese rats and a decrease in the expression of the ACE2-AT2R-MasR axis in obese Zucker rats. In a study of a left nephrectomy rat model[20], HS diet intake increased the glomerular ACE/ACE2 ratio, which was associated with decreased ACE2. In ACE2 KO mice, ACE2 deficiency significantly increased renal oxidative stress by reducing the production of Ang 1-7. In another study, the consumption of a HS diet in normotensive animals reduced ACE2 protein expression and raised renal oxidative stress[21]. Another study showed that the ACE/ACE2 protein ratio was increased in the kidneys of spontaneously hypertensive rats (SHRs) when fed diets with high levels of NaCl. At the same time, compared to the other SHR groups, there was an increase in kidney ACE2 protein and activity in the group fed a long-term low salt diet[18]. Similarly, Varagic *et al*[22] used an SHR model to prove that HS intake decreased cardiac ACE2 mRNA and protein expression. In Dahl salt-sensitive hypertensive rats, a HS diet reduced ACE2 mRNA expression and augmented the local RAS, which induced hypertension[23]. The HS diet also decreased ACE2 protein expression as assessed using immunohistochemistry compared to a normal-salt diet[24].

In summary, these findings show that a HS diet can reduce the expression of ACE2, thereby affecting the RAS. Further studies have reported that interventions that augment ACE2 expression or activity can be helpful to prevent cardiovascular damage[25]. Therefore, elucidating the precise mechanisms involved in the interaction of sodium intake and ACE2 will be conducive to predicting the physiological and pathological changes caused by a HS diet (Table 1).

**ACE2 and RAS inhibitors**

Some studies in animal models have demonstrated that both ACEIs (lisinopril, enalapril, and ramipril) and ARBs (losartan, olmesartan, and telmisartan) can upregulate ACE2 expression. Ferrario *et al*[26] showed that *ACE2* mRNA expression increased in the left ventricle of normotensive Lewis rats after 12 d of lisinopril or losartan treatment. Lisinopril increased ACE2 levels by 5-fold, while losartan increased ACE2 levels by 3-fold. A rat study from Ocaranza *et al*[27] showed that enalapril prevented the decrease in mRNA levels and activities of ACE2 in late ventricular dysfunction after myocardial infarction. The results of Ishiyama *et al*[28] indicated that the level of *ACE2* mRNA increased after ARB treatment in rats with coronary artery ligation. A mouse study by Soler *et al*[29] showed that ACE2 is preferentially localized in the tunica media of kidney arterioles, and its expression is amplified after administration of telmisartan. On the other hand, some association studies have shown no increase in *ACE2* mRNA after ACEI or ARB treatment. An animal study by Burrell *et al*[30] found that ramipril attenuated cardiac hypertrophy and inhibited cardiac ACE but had no effect on cardiac *ACE2* mRNA in rats after coronary artery ligation. Subsequent studies by Burchill *et al*[31] provided evidence that there was no increase in ACE2 mRNA or protein expression in rats after coronary artery ligation and treatment with valsartan, ramipril, or both when compared to the control group. In general, these studies on experimental animals did not provide consistent evidence to prove the effect of ARB/ACEI administration on ACE2 protein expression. In the case of simulating human drug delivery, further experimental studies are needed.

With the pandemic of COVID-19 spreading since January 2020, it has been inferred that increased ACE2 expression in the lungs correlated with a higher risk of SARS-CoV-2 infection in patients with cardiac and renal diseases, hypertension, and diabetes treated with ACEIs or ARBs. However, a review from Sriram and Insel[32] showed that there was no clear evidence that elevated ACE2 expression when using ACEIs/ARBs could increase the risk of SARS-CoV-2 infection. In a study of 362 hospitalized hypertensive COVID-19 patients, there was no significant difference between severe and noncritical patients or between non-survivors and survivors according to the use of ACEIs and/or ARBs[33]. Conversely, in a recent meta-analysis from Chu *et al*[34], it was suggested that ACEI treatment reduced the risk of infection with SARS-CoV-2 and that blocking the RAS might decrease all-cause mortality in COVID-19 patients. This study also reported that ACEIs reduced the risk of non-COVID pneumonia and all-cause mortality caused by non-COVID pneumonia. The effect of ACEIs/ARBs on the expression of ACE2 and the effect on human infection with SARS-CoV-2 remain unknown and complex. Some research has supported the hypothesis that ACE inhibition by ACEIs might stimulate negative feedback, upregulating ACE2 expression but decreasing overall inflammation in the absence of angiotensin II[35]. Thus, the effects of ACEIs/ARBs vary depending on the clinical stage: Negative in the initial infection phase but positive in the tissue inflammation stage[36,37]. Most researchers believe that the use of ARB or ACEI drugs should not be stopped for the purpose of reducing SARS-CoV-2 infection. Stopping maintenance treatment may cause blood pressure imbalance or HF. The guidelines should be revised quickly based on various clinical data, and personalized treatment should be carried out in accordance with clinical manifestations[38] (Table 2).

**ACE2 and diseases**

The functional components of the RAS are in balance with each other to maintain the health of the body. Under certain pathological conditions, Ang II, AT1R, or ACE levels can increase or become unbalanced, which is detrimental. It has been two decades since the discovery of ACE2. During this period, efforts towards the characterization of this enzyme have provided greater insights into the RAS. The ongoing studies provoked more questions, particularly regarding the role of ACE2 in the development and progression of hypertension and renal injury, as well as other pathologies, including diabetes, HF, liver fibrosis, and lung injury (Tables 3 and 4).

***ACE2 and its role in the kidney and diabetes***

The kidney possesses a fully functional local RAS capable of producing Ang II, a major contributor to the progression of chronic kidney disease. ACE2 is highly expressed in the kidney and predominantly localized to proximal tubules and glomerular podocytes[39,40]. Several lines of evidence indicate that ACE2 serves as a key protective enzyme to prevent progressive renal damage by reducing oxidative stress, inflammation, and fibrosis[41-43].

The study of ACE2 in the context of diabetes has focused primarily on the kidney. ACE2 may be an important target for the treatment and prevention of diabetic nephropathy (DN). ACE2 expression in the kidney has been studied in both type 1 diabetes (T1D) and type 2 diabetes (T2D) models. The majority of animal studies indicate that ACE2 expression is downregulated in the glomeruli in diabetes, whereas tubular ACE2 expression is upregulated[41,44-46]. Tikellis *et al*[47] first reported that ACE2 expression was reduced in the kidneys of rats with longstanding diabetes mellitus. In 8-wk-old db/db mice, a model of early T2D, ACE2 expression is elevated, while ACE expression is decreased in both glomeruli and the cortex[48] prior to the development of DN. In another study of db/db mice, Chodavarapu *et al*[45] demonstrated that the protein expression of ACE2 was reduced in glomeruli, while tubular ACE2 and a disintegrin and metalloprotease 17 were increased. In two models of T1D [streptozotocin (STZ)-induced and Akita mouse (Ins2WT/C96Y) models], *ACE2* gene deletion accelerated the development of DN[49,50], which could be ameliorated by perindopril or irbesartan. Moreover, treatment with recombinant human ACE2 (rhACE2) in male Akita mice led to reductions in albuminuria, hypertension, plasma Ang II levels, activation of NADPH oxidase, glomerular hypertrophy, and mesangial matrix expansion, thereby preventing the progression of DN[51]. In another rat study, the injection of adenoviral (Ad)-ACE2 in STZ-induced diabetic rats for 4 wk improved many signs of DN[52]. Furthermore, Ad-ACE2 and ACEI had similar effects, whereas the combined use of Ad-ACE2 and ACEI offered no additional benefits[52].

In humans, the expression of ACE2 was significantly reduced in both the glomeruli and proximal tubules in biopsy samples collected from patients with T2D-induced kidney disease[53]. Conversely, in a real-time polymerase chain reaction study, *ACE2* mRNA expression was not significantly changed in eight diabetic patients with overt proteinuria compared with 66 nondiabetic patients with renal disease[54]. The differences in the results obtained in human studies of T2D nephropathy might be due to the different stages of diabetes. To date, no human studies of early-stage diabetes have been conducted. Moreover, genetic variation in and around the gene encoding ACE2 is most often detected using single nucleotide polymorphisms (SNPs). In a recent study, 14 *ACE2* polymorphisms were genotyped by matrix-assisted laser desorption ionization time-of-flight mass spectrometry in the Uygur population of the Xinjiang region of China. Among them, the *ACE2* SNPs rs2074192, rs4240157, rs4646188, and 879922 were associated with increased microalbuminuria in T2D patients[55].

Taken together, the above studies suggest that ACE2 might play a protective role against the development of DN.

***ACE2 and hypertension***

The role of ACE2 has been intensively studied in models of hypertension. Crackower *et al[*56] first reported that ACE2 transgenic mice exhibited lower blood pressure than wild-type mice. Subsequent studies reported that ACE2 probably has a small effect on blood pressure in mice under normal conditions[57-59]. However, it plays a much more prominent role in the regulation of hypertension, especially when Ang II levels are elevated. Existing studies have shown that ACE2 was reduced in kidneys from rat models of hypertension, such as salt-sensitive Sabra hypertensive rats, SHRs, and stroke-prone SHRs (SHRSP)[56,60]. Moreover, in a sheep model of fetal programmed hypertension, the administration of betamethasone on the 80th day of gestation markedly reduced ACE2 activity in the proximal tubules and urine in adolescent sheep[61]. Rentzsch *et al*[62] assessed the role of ACE2 in the pathogenesis of hypertension. These authors generated transgenic rats in a SHRSP genetic background, called SHRSP-ACE2, which expressed human ACE2 in vascular smooth muscle cells under the control of the smooth muscle 22α promoter. They found that endothelial function was significantly improved in SHRSP-ACE2 rats compared with SHRSP rats. These data indicate that vascular ACE2 overexpression in SHRSP reduces hypertension, probably through local Ang II degradation and by improving endothelial function. Existing animal studies have shown that the administration of recombinant ACE2 (rACE2) degrades Ang II, lowers blood pressure, and attenuates Ang II–induced organ injury. One study showed that in the SHR model, rhACE2 partly corrected hypertension and NADPH oxidase activation and increased superoxide generation in the heart, kidney, and blood vessels over a 14-d period[63]. Moreover, the prevention of Ang II-induced hypertension by mouse rACE2 was completely abolished by the specific ACE2 inhibitor MLN-4760, a nonpeptide inhibitor no longer available from Millennium Pharmaceuticals[64].

In human studies, Patel *et al*[65] found that in Caucasians with T2D, genetic variation in *ACE2* is associated with hypertension and reduced systolic function in men and hypertension and increased left ventricular mass in women. Liu *et al*[55] found that the *ACE2* SNPs rs2048683, rs233575, rs4240157, rs4646156, rs4646188, and rs879922 were associated with increased systolic blood pressure (SBP), while rs2074192, rs4646188, and rs879922 were associated with elevated diastolic blood pressure in Uygur T2D patients. Luo *et al*[66] revealed that the *ACE2* variant rs2074192 was associated with essential hypertension (EH), while three *ACE2* variants (rs4240157, rs4646155, and rs4830542) were associated with EH- and hypertension-related atrial fibrillation and left atrial remodeling in south Xinjiang, China. In another study, the *ACE2* rs2106809 T allele was found to confer a 1.6-fold risk for hypertension in women[67]. Additionally, one study showed that aberrant methylation of the *ACE2* promoter may be associated with EH risk[68].

These findings indicate that ACE2 is a key regulator that maintains the balance of blood pressure. In animal studies, it has been demonstrated that the administration of recombinant ACE2 has a beneficial effect on the treatment of hypertension. Currently, large clinical trials to explore this and related alternative interventions are underway.

***ACE2 and cardiac function, ventricular remodeling, and HF***

SARS-CoV-2 enters the upper respiratory epithelium and lungs predominantly through ACE2. Nevertheless, in a single-center report of 416 patients hospitalized with COVID-19, 19.7% showed evidence of cardiac injury, suggesting a possible pathologic role for myocardial ACE2 expression[69]. ACE2 is present on endothelial cells and can undergo so-called shedding into the circulation. In patients with cardiovascular disease, increased ACE2 activity in the circulation predicts adverse cardiovascular outcomes in patients with HF, coronary artery disease, and aortic stenosis[70].

Crackower *et al*[56] demonstrated the first evidence that ACE2 may have a role in cardiac function. They found that ACE2 deletion in mice resulted in a severe heart contractility defect, increased levels of Ang II in the kidney, heart, and plasma, and upregulation of hypoxia-induced genes in the heart. Conversely, Gurley *et al*[57] reported that ACE2 deletion enhanced susceptibility to Ang II-induced hypertension but had no effect on cardiac structure or function. Huentelman *et al*[71] showed that ACE2 overexpression protects the heart from Ang II-induced hypertrophy and fibrosis. Another study on SHR hypertensive rats also showed that ACE2 overexpression exerted protective effects against high blood pressure and cardiac pathophysiology induced by hypertension[72]. Similarly, in a rabbit atherosclerosis model, local overexpression of ACE2 significantly inhibited the development of early atherosclerotic lesions[73].

In humans, increased circulating ACE2 activity is associated with coronary heart disease and HF, and a large proportion of the variation in plasma ACE2 levels is attributed to hereditary factors. One study showed that ACE2 activity was significantly increased in HF patients with reduced ejection fraction (HFrEF). Serum ACE2 activity was negatively correlated with left ventricular systolic function in HfrEF[74]. In addition, one study measured soluble ACE2 (sACE2) activity in 113 patients with chronic systolic HF and showed that elevated plasma sACE2 activity was associated with greater severity of myocardial dysfunction, which indicated that plasma sACE2 activity might be an independent predictor of adverse clinical events[75]. A recent study published in Lancet presented one of the largest epidemiological datasets on plasma ACE2 concentration in the general population[76]. They performed a case-cohort study involving 10753 participants from the multinational Prospective Urban Rural Epidemiology study, including 5084 patients randomly selected as the sub-cohort and 5669 with an incident event of interest. They reported that ACE2 concentration was the highest-ranked independent predictor of death compared with standard cardiovascular risk markers (smoking, diabetes, SBP, non-high density lipoprotein cholesterol, and body mass index). An increased concentration of plasma ACE2 was associated with an increased risk of all-cause mortality [hazard ratio (HR): 1.35 per 1 standard deviation (SD) increase; 95% confidence interval (CI): 1.29-1.43], incident HF (HR: 1.27 per 1 SD increase; 95%CI: 1.10-1.46), stroke (HR: 1.21 per 1 SD increase; 95%CI: 1.10-1.32), myocardial infarction (HR: 1.23 per 1 SD increase; 95%CI: 1.13-1.33), and incident diabetes (HR: 1.44 per 1 SD increase; 95%CI: 1.36-1.52). Other studies have investigated whether the *ACE2* gene is associated with left ventricular hypertrophy and coronary artery disease. The *ACE2* SNPs most frequently used in association studies are rs2285666 and rs1978124[77]. Recently, Liu *et al*[55] found that the *ACE2* SNPs rs2074192 and rs879922 were associated with carotid arteriosclerosis stenosis and that the *ACE2* SNPs rs2048683, rs4240157, rs4646156, rs4646188, and rs879922 were linked to more substantial left heart remodeling. Furthermore, one study assessed ACE2 expression by performing bulk and single nucleus RNA-Seq on the left ventricles of 11 individuals with dilated cardiomyopathy, 15 individuals with hypertrophic cardiomyopathy, and 16 controls with nonfailing hearts from the Penn Human Heart Tissue Biobank. They found that cardiac ACE2 expression was down-regulated in fibroblasts, pericytes, and vascular smooth muscle but upregulated in cardiomyocytes[69].

Investigations of ACE2 as well as its role in cardiac function and HF will undoubtedly provide greater insight into the roles of this enzyme. However, carefully conducted large-scale clinical studies are urgently needed to clarify the potential role of ACE2 in cardiovascular diseases more precisely.

***ACE2 and acute lung injury***

Like many other organ lung cells also have a local RAS[78], which influences the pathogenesis of lung injury *via* cellular effects, including changes in vascular permeability, vascular tone, fibroblast activity, or alveolar epithelial cell apoptosis[79,80]. ACE2 plays a pivotal role in Ang II degradation in the RAS cascade and thus limits inflammation and fibrosis in the lung[81]. Imai *et al*[82] investigated the role of ACE2 in acute respiratory distress syndrome (ARDS) by using ACE2 knockout mice. In three different ARDS models (acid-aspiration-induced, endotoxin-induced, and peritoneal sepsis-induced ARDS), it was shown that a loss of ACE2 expression in mutant mice resulted in enhanced vascular permeability, increased lung edema, engendered neutrophil accumulation, and worsened lung function. Importantly, treatment with catalytically active recombinant ACE2 protein improved the symptoms of acute lung injury in both wild-type mice and ACE2 knockout mice. Thus, ACE2 plays a protective role in acute lung injury. Mechanically, the finding that reduced ACE2 on lung cell surfaces is correlated with lung damage due to an uncontrolled RAS cascade is supported by data about the effects of long-lasting hyperoxia on pulmonary tissue[82-86]. In addition, a study revealed that ACE2 activation can reduce the severity of lipopolysaccharide-induced acute lung injury *via* the activated serine/threonine protein kinase/mammalian target of rapamycin pathway[87].

In the current pandemic of COVID-19, both bioinformatics modeling and *in vitro* experiments indicate that SARS-CoV-2 likely utilizes ACE2 as a receptor to gain entry into human cells[88-90]. A recent study evaluated lung function in a mouse model of SARS-CoV-2 infection. They used transgenic mice expressing the human ACE2 (hACE2) receptor driven by the cytokeratin-18 (*K18*) gene promoter (K18-hACE2) and found that intranasal inoculation of SARS-CoV-2 in K18-hACE2 mice resulted in high levels of viral infection in the lungs, with spread to other organs[91]. Similarly, one study also used a model of mice expressing hACE2 in the lung. In this study, the mice were transduced by oropharyngeal delivery of recombinant human adenovirus type 5 expressing hACE2[92]. They found that mice were infected with SARS-CoV-2 at day 4 post-transduction and developed interstitial pneumonia related to perivascular inflammation. On the other hand, as described above, a similar decrease in ACE2 has also been seen in cases of COVID-19 with severe lung injury, which might be attributable to the negative consequences that arise from insufficient Ang II degradation[93]. In human trials, GSK2586881, a rhACE2, was well tolerated in 44 patients with ARDS and has been found to reduce Ang II levels and increase Ang 1-7 levels, although it failed to improve the physiological and clinical indicators of ARDS in patients[94]. This study likely represents the first clinical application of rhACE2 in the field of ARDS, so we speculate that rhACE2 may become one of the most promising approaches for protecting against lung injury in patients with COVID-19.

Based on the above description, ACE2 plays a complex role in COVID-19-induced acute lung injury. On the one hand, high levels of ACE2 receptors on the cell surface may accelerate the invasion of SARS-CoV-2 during the very early phase of infection. On the other hand, low levels of ACE2 can ultimately worsen the disease course due to insufficient Ang II conversion in cases of severe COVID-19 with pulmonary complications.

**CONCLUSION**

As a vital component of the RAS, ACE2 is closely related to the occurrence and development of RAS-associated diseases. A better understanding of the biological functions of ACE2 will be beneficial to the treatment. Previous studies show that a high-salt diet can decrease the expression of ACE2 and cause RAS disorders. Recent reports have not yet indicated that ARBs or ACEIs will increase the level of ACE2, thereby aggravating SARS-CoV-2 infection. Therefore, the mechanisms involved need to be further improved. As ACE2 is an important receptor through which SARS-CoV-2 can invade cells, further studies on ACE2 should focus on the development of drugs that inhibit the virus from entering cells and impede the binding of the S protein to ACE2 for COVID-19 treatment. More research on ACE2 should be conducted in the future to carry out targeted and effective treatment at a higher level.

**REFERENCES**

1 **Li W**, Moore MJ, Vasilieva N, Sui J, Wong SK, Berne MA, Somasundaran M, Sullivan JL, Luzuriaga K, Greenough TC, Choe H, Farzan M. Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. *Nature* 2003; **426**: 450-454 [PMID: 14647384 DOI: 10.1038/nature02145]

2 **Fang L**, Karakiulakis G, Roth M. Are patients with hypertension and diabetes mellitus at increased risk for COVID-19 infection? *Lancet Respir Med* 2020; **8**: e21 [PMID: 32171062 DOI: 10.1016/S2213-2600(20)30116-8]

3 **Zheng YY**, Ma YT, Zhang JY, Xie X. COVID-19 and the cardiovascular system. *Nat Rev Cardiol* 2020; **17**: 259-260 [PMID: 32139904 DOI: 10.1038/s41569-020-0360-5]

4 **Yan R**, Zhang Y, Li Y, Xia L, Guo Y, Zhou Q. Structural basis for the recognition of SARS-CoV-2 by full-length human ACE2. *Science* 2020; **367**: 1444-1448 [PMID: 32132184 DOI: 10.1126/science.abb2762]

5 **Tipnis SR**, Hooper NM, Hyde R, Karran E, Christie G, Turner AJ. A human homolog of angiotensin-converting enzyme. Cloning and functional expression as a captopril-insensitive carboxypeptidase. *J Biol Chem* 2000; **275**: 33238-33243 [PMID: 10924499 DOI: 10.1074/jbc.M002615200]

6 **Donoghue M**, Hsieh F, Baronas E, Godbout K, Gosselin M, Stagliano N, Donovan M, Woolf B, Robison K, Jeyaseelan R, Breitbart RE, Acton S. A novel angiotensin-converting enzyme-related carboxypeptidase (ACE2) converts angiotensin I to angiotensin 1-9. *Circ Res* 2000; **87**: E1-E9 [PMID: 10969042 DOI: 10.1161/01.res.87.5.e1]

7 **Castardeli C**, Sartório CL, Pimentel EB, Forechi L, Mill JG. The ACE 2 activator diminazene aceturate (DIZE) improves left ventricular diastolic dysfunction following myocardial infarction in rats. *Biomed Pharmacother* 2018; **107**: 212-218 [PMID: 30092400 DOI: 10.1016/j.biopha.2018.07.170]

8 **Bradford CN**, Ely DR, Raizada MK. Targeting the vasoprotective axis of the renin-angiotensin system: a novel strategic approach to pulmonary hypertensive therapy. *Curr Hypertens Rep* 2010; **12**: 212-219 [PMID: 20556668 DOI: 10.1007/s11906-010-0122-6]

9 **Turner AJ**, Hooper NM. The angiotensin-converting enzyme gene family: genomics and pharmacology. *Trends Pharmacol Sci* 2002; **23**: 177-183 [PMID: 11931993 DOI: 10.1016/s0165-6147(00)01994-5]

10 **Turner AJ**, Hiscox JA, Hooper NM. ACE2: from vasopeptidase to SARS virus receptor. *Trends Pharmacol Sci* 2004; **25**: 291-294 [PMID: 15165741 DOI: 10.1016/j.tips.2004.04.001]

11 **Passos-Silva DG**, Verano-Braga T, Santos RA. Angiotensin-(1-7): beyond the cardio-renal actions. *Clin Sci (Lond)* 2013; **124**: 443-456 [PMID: 23249272 DOI: 10.1042/CS20120461]

12 **Zhang YH**, Zhang YH, Dong XF, Hao QQ, Zhou XM, Yu QT, Li SY, Chen X, Tengbeh AF, Dong B, Zhang Y. ACE2 and Ang-(1-7) protect endothelial cell function and prevent early atherosclerosis by inhibiting inflammatory response. *Inflamm Res* 2015; **64**: 253-260 [PMID: 25721616 DOI: 10.1007/s00011-015-0805-1]

13 **Lovren F**, Pan Y, Quan A, Teoh H, Wang G, Shukla PC, Levitt KS, Oudit GY, Al-Omran M, Stewart DJ, Slutsky AS, Peterson MD, Backx PH, Penninger JM, Verma S. Angiotensin converting enzyme-2 confers endothelial protection and attenuates atherosclerosis. *Am J Physiol Heart Circ Physiol* 2008; **295**: H1377-H1384 [PMID: 18660448 DOI: 10.1152/ajpheart.00331.2008]

14 **Wang Y**, Tikellis C, Thomas MC, Golledge J. Angiotensin converting enzyme 2 and atherosclerosis. *Atherosclerosis* 2013; **226**: 3-8 [PMID: 22947420 DOI: 10.1016/j.atherosclerosis.2012.08.018]

15 **Fraga-Silva RA**, Da Silva DG, Montecucco F, Mach F, Stergiopulos N, da Silva RF, Santos RA. The angiotensin-converting enzyme 2/angiotensin-(1-7)/Mas receptor axis: a potential target for treating thrombotic diseases. *Thromb Haemost* 2012; **108**: 1089-1096 [PMID: 23093373 DOI: 10.1160/TH12-06-0396]

16 **Lubel JS**, Herath CB, Tchongue J, Grace J, Jia Z, Spencer K, Casley D, Crowley P, Sievert W, Burrell LM, Angus PW. Angiotensin-(1-7), an alternative metabolite of the renin-angiotensin system, is up-regulated in human liver disease and has antifibrotic activity in the bile-duct-ligated rat. *Clin Sci (Lond)* 2009; **117**: 375-386 [PMID: 19371232 DOI: 10.1042/CS20080647]

17 **Kuba K**, Imai Y, Penninger JM. Multiple functions of angiotensin-converting enzyme 2 and its relevance in cardiovascular diseases. *Circ J* 2013; **77**: 301-308 [PMID: 23328447 DOI: 10.1253/circj.cj-12-1544]

18 **Berger RC**, Vassallo PF, Crajoinas Rde O, Oliveira ML, Martins FL, Nogueira BV, Motta-Santos D, Araújo IB, Forechi L, Girardi AC, Santos RA, Mill JG. Renal Effects and Underlying Molecular Mechanisms of Long-Term Salt Content Diets in Spontaneously Hypertensive Rats. *PLoS One* 2015; **10**: e0141288 [PMID: 26495970 DOI: 10.1371/journal.pone.0141288]

19 **Samuel P**, Ali Q, Sabuhi R, Wu Y, Hussain T. High Na intake increases renal angiotensin II levels and reduces expression of the ACE2-AT(2)R-MasR axis in obese Zucker rats. *Am J Physiol Renal Physiol* 2012; **303**: F412-F419 [PMID: 22592638 DOI: 10.1152/ajprenal.00097.2012]

20 **Bernardi S**, Toffoli B, Zennaro C, Tikellis C, Monticone S, Losurdo P, Bellini G, Thomas MC, Fallo F, Veglio F, Johnston CI, Fabris B. High-salt diet increases glomerular ACE/ACE2 ratio leading to oxidative stress and kidney damage. *Nephrol Dial Transplant* 2012; **27**: 1793-1800 [PMID: 22036945 DOI: 10.1093/ndt/gfr600]

21 **Shimoura CG**, Lincevicius GS, Nishi EE, Girardi AC, Simon KA, Bergamaschi CT, Campos RR. Increased Dietary Salt Changes Baroreceptor Sensitivity and Intrarenal Renin-Angiotensin System in Goldblatt Hypertension. *Am J Hypertens* 2017; **30**: 28-36 [PMID: 27629265 DOI: 10.1093/ajh/hpw107]

22 **Varagic J**, Ahmad S, Brosnihan KB, Groban L, Chappell MC, Tallant EA, Gallagher PE, Ferrario CM. Decreased cardiac Ang-(1-7) is associated with salt-induced cardiac remodeling and dysfunction. *Ther Adv Cardiovasc Dis* 2010; **4**: 17-25 [PMID: 19946038 DOI: 10.1177/1753944709353337]

23 **Takeda Y**, Zhu A, Yoneda T, Usukura M, Takata H, Yamagishi M. Effects of aldosterone and angiotensin II receptor blockade on cardiac angiotensinogen and angiotensin-converting enzyme 2 expression in Dahl salt-sensitive hypertensive rats. *Am J Hypertens* 2007; **20**: 1119-1124 [PMID: 17903697 DOI: 10.1016/j.amjhyper.2007.05.008]

24 **Cao G**, Della Penna SL, Kouyoumdzian NM, Choi MR, Gorzalczany S, Fernández BE, Toblli JE, Rosón MI. Immunohistochemical expression of intrarenal renin angiotensin system components in response to tempol in rats fed a high salt diet. *World J Nephrol* 2017; **6**: 29-40 [PMID: 28101449 DOI: 10.5527/wjn.v6.i1.29]

25 **Tikellis C**, Bernardi S, Burns WC. Angiotensin-converting enzyme 2 is a key modulator of the renin-angiotensin system in cardiovascular and renal disease. *Curr Opin Nephrol Hypertens* 2011; **20**: 62-68 [PMID: 21099686 DOI: 10.1097/MNH.0b013e328341164a]

26 **Ferrario CM**, Jessup J, Chappell MC, Averill DB, Brosnihan KB, Tallant EA, Diz DI, Gallagher PE. Effect of angiotensin-converting enzyme inhibition and angiotensin II receptor blockers on cardiac angiotensin-converting enzyme 2. *Circulation* 2005; **111**: 2605-2610 [PMID: 15897343 DOI: 10.1161/CIRCULATIONAHA.104.510461]

27 **Ocaranza MP**, Godoy I, Jalil JE, Varas M, Collantes P, Pinto M, Roman M, Ramirez C, Copaja M, Diaz-Araya G, Castro P, Lavandero S. Enalapril attenuates downregulation of Angiotensin-converting enzyme 2 in the late phase of ventricular dysfunction in myocardial infarcted rat. *Hypertension* 2006; **48**: 572-578 [PMID: 16908757 DOI: 10.1161/01.HYP.0000237862.94083.45]

28 **Ishiyama Y**, Gallagher PE, Averill DB, Tallant EA, Brosnihan KB, Ferrario CM. Upregulation of angiotensin-converting enzyme 2 after myocardial infarction by blockade of angiotensin II receptors. *Hypertension* 2004; **43**: 970-976 [PMID: 15007027 DOI: 10.1161/01.HYP.0000124667.34652.1a]

29 **Soler MJ**, Ye M, Wysocki J, William J, Lloveras J, Batlle D. Localization of ACE2 in the renal vasculature: amplification by angiotensin II type 1 receptor blockade using telmisartan. *Am J Physiol Renal Physiol* 2009; **296**: F398-F405 [PMID: 19004932 DOI: 10.1152/ajprenal.90488.2008]

30 **Burrell LM**, Risvanis J, Kubota E, Dean RG, MacDonald PS, Lu S, Tikellis C, Grant SL, Lew RA, Smith AI, Cooper ME, Johnston CI. Myocardial infarction increases ACE2 expression in rat and humans. *Eur Heart J* 2005; **26**: 369-75; discussion 322-4 [PMID: 15671045 DOI: 10.1093/eurheartj/ehi114]

31 **Burchill LJ**, Velkoska E, Dean RG, Griggs K, Patel SK, Burrell LM. Combination renin-angiotensin system blockade and angiotensin-converting enzyme 2 in experimental myocardial infarction: implications for future therapeutic directions. *Clin Sci (Lond)* 2012; **123**: 649-658 [PMID: 22715807 DOI: 10.1042/CS20120162]

32 **Sriram K**, Insel PA. Risks of ACE Inhibitor and ARB Usage in COVID-19: Evaluating the Evidence. *Clin Pharmacol Ther* 2020; **108**: 236-241 [PMID: 32320478 DOI: 10.1002/cpt.1863]

33 **Li J**, Wang X, Chen J, Zhang H, Deng A. Association of Renin-Angiotensin System Inhibitors With Severity or Risk of Death in Patients With Hypertension Hospitalized for Coronavirus Disease 2019 (COVID-19) Infection in Wuhan, China. *JAMA Cardiol* 2020; **5**: 825-830 [PMID: 32324209 DOI: 10.1001/jamacardio.2020.1624]

34 **Chu C**, Zeng S, Hasan AA, Hocher CF, Krämer BK, Hocher B. Comparison of infection risks and clinical outcomes in patients with and without SARS-CoV-2 lung infection under renin-angiotensin-aldosterone system blockade: Systematic review and meta-analysis. *Br J Clin Pharmacol* 2020 [PMID: 33217033 DOI: 10.1111/bcp.14660]

35 **Li XC**, Zhang J, Zhuo JL. The vasoprotective axes of the renin-angiotensin system: Physiological relevance and therapeutic implications in cardiovascular, hypertensive and kidney diseases. *Pharmacol Res* 2017; **125**: 21-38 [PMID: 28619367 DOI: 10.1016/j.phrs.2017.06.005]

36 **Sommerstein R**, Kochen MM, Messerli FH, Gräni C. Coronavirus Disease 2019 (COVID-19): Do Angiotensin-Converting Enzyme Inhibitors/Angiotensin Receptor Blockers Have a Biphasic Effect? *J Am Heart Assoc* 2020; **9**: e016509 [PMID: 32233753 DOI: 10.1161/JAHA.120.016509]

37 **Guo J**, Huang Z, Lin L, Lv J. Coronavirus Disease 2019 (COVID-19) and Cardiovascular Disease: A Viewpoint on the Potential Influence of Angiotensin-Converting Enzyme Inhibitors/Angiotensin Receptor Blockers on Onset and Severity of Severe Acute Respiratory Syndrome Coronavirus 2 Infection. *J Am Heart Assoc* 2020; **9**: e016219 [PMID: 32233755 DOI: 10.1161/JAHA.120.016219]

38 **Alexandre J**, Cracowski JL, Richard V, Bouhanick B; 'Drugs, COVID-19' working group of the French Society of Pharmacology, Therapeutics. Renin-angiotensin-aldosterone system and COVID-19 infection. *Ann Endocrinol (Paris)* 2020; **81**: 63-67 [PMID: 32370986 DOI: 10.1016/j.ando.2020.04.005]

39 **Warner FJ**, Guy JL, Lambert DW, Hooper NM, Turner AJ. Angiotensin Converting Enzyme-2 (ACE2) and its Possible Roles in Hypertension, Diabetes and Cardiac Function. *Lett Pept Sci* 2003; **10**: 377-385 [PMID: 32214681 DOI: 10.1007/s10989-004-2387-6]

40 **Mizuiri S**, Ohashi Y. ACE and ACE2 in kidney disease. *World J Nephrol* 2015; **4**: 74-82 [PMID: 25664248 DOI: 10.5527/wjn.v4.i1.74]

41 **Batlle D**, Jose Soler M, Ye M. ACE2 and diabetes: ACE of ACEs? *Diabetes* 2010; **59**: 2994-2996 [PMID: 21115782 DOI: 10.2337/db10-1205]

42 **Bindom SM**, Lazartigues E. The sweeter side of ACE2: physiological evidence for a role in diabetes. *Mol Cell Endocrinol* 2009; **302**: 193-202 [PMID: 18948167 DOI: 10.1016/j.mce.2008.09.020]

43 **Rao S**, Lau A, So HC. Exploring Diseases/Traits and Blood Proteins Causally Related to Expression of ACE2, the Putative Receptor of SARS-CoV-2: A Mendelian Randomization Analysis Highlights Tentative Relevance of Diabetes-Related Traits. *Diabetes Care* 2020; **43**: 1416-1426 [PMID: 32430459 DOI: 10.2337/dc20-0643]

44 **Goru SK**, Kadakol A, Malek V, Pandey A, Sharma N, Gaikwad AB. Diminazene aceturate prevents nephropathy by increasing glomerular ACE2 and AT2 receptor expression in a rat model of type1 diabetes. *Br J Pharmacol* 2017; **174**: 3118-3130 [PMID: 28688122 DOI: 10.1111/bph.13946]

45 **Chodavarapu H**, Grobe N, Somineni HK, Salem ES, Madhu M, Elased KM. Rosiglitazone treatment of type 2 diabetic db/db mice attenuates urinary albumin and angiotensin converting enzyme 2 excretion. *PLoS One* 2013; **8**: e62833 [PMID: 23646149 DOI: 10.1371/journal.pone.0062833]

46 **Ye M**, Wysocki J, William J, Soler MJ, Cokic I, Batlle D. Glomerular localization and expression of Angiotensin-converting enzyme 2 and Angiotensin-converting enzyme: implications for albuminuria in diabetes. *J Am Soc Nephrol* 2006; **17**: 3067-3075 [PMID: 17021266 DOI: 10.1681/ASN.2006050423]

47 **Tikellis C**, Johnston CI, Forbes JM, Burns WC, Burrell LM, Risvanis J, Cooper ME. Characterization of renal angiotensin-converting enzyme 2 in diabetic nephropathy. *Hypertension* 2003; **41**: 392-397 [PMID: 12623933 DOI: 10.1161/01.HYP.0000060689.38912.CB]

48 **Ye M**, Wysocki J, Naaz P, Salabat MR, LaPointe MS, Batlle D. Increased ACE 2 and decreased ACE protein in renal tubules from diabetic mice: a renoprotective combination? *Hypertension* 2004; **43**: 1120-1125 [PMID: 15078862 DOI: 10.1161/01.HYP.0000126192.27644.76]

49 **Wong DW**, Oudit GY, Reich H, Kassiri Z, Zhou J, Liu QC, Backx PH, Penninger JM, Herzenberg AM, Scholey JW. Loss of angiotensin-converting enzyme-2 (Ace2) accelerates diabetic kidney injury. *Am J Pathol* 2007; **171**: 438-451 [PMID: 17600118 DOI: 10.2353/ajpath.2007.060977]

50 **Tikellis C**, Bialkowski K, Pete J, Sheehy K, Su Q, Johnston C, Cooper ME, Thomas MC. ACE2 deficiency modifies renoprotection afforded by ACE inhibition in experimental diabetes. *Diabetes* 2008; **57**: 1018-1025 [PMID: 18235039 DOI: 10.2337/db07-1212]

51 **Oudit GY**, Liu GC, Zhong J, Basu R, Chow FL, Zhou J, Loibner H, Janzek E, Schuster M, Penninger JM, Herzenberg AM, Kassiri Z, Scholey JW. Human recombinant ACE2 reduces the progression of diabetic nephropathy. *Diabetes* 2010; **59**: 529-538 [PMID: 19934006 DOI: 10.2337/db09-1218]

52 **Liu CX**, Hu Q, Wang Y, Zhang W, Ma ZY, Feng JB, Wang R, Wang XP, Dong B, Gao F, Zhang MX, Zhang Y. Angiotensin-converting enzyme (ACE) 2 overexpression ameliorates glomerular injury in a rat model of diabetic nephropathy: a comparison with ACE inhibition. *Mol Med* 2011; **17**: 59-69 [PMID: 20844835 DOI: 10.2119/molmed.2010.00111]

53 **Reich HN**, Oudit GY, Penninger JM, Scholey JW, Herzenberg AM. Decreased glomerular and tubular expression of ACE2 in patients with type 2 diabetes and kidney disease. *Kidney Int* 2008; **74**: 1610-1616 [PMID: 19034303 DOI: 10.1038/ki.2008.497]

54 **Konoshita T**, Wakahara S, Mizuno S, Motomura M, Aoyama C, Makino Y, Kawai Y, Kato N, Koni I, Miyamori I, Mabuchi H. Tissue gene expression of renin-angiotensin system in human type 2 diabetic nephropathy. *Diabetes Care* 2006; **29**: 848-852 [PMID: 16567826 DOI: 10.2337/diacare.29.04.06.dc05-1873]

55 **Liu C**, Li Y, Guan T, Lai Y, Shen Y, Zeyaweiding A, Zhao H, Li F, Maimaiti T. ACE2 polymorphisms associated with cardiovascular risk in Uygurs with type 2 diabetes mellitus. *Cardiovasc Diabetol* 2018; **17**: 127 [PMID: 30227878 DOI: 10.1186/s12933-018-0771-3]

56 **Crackower MA**, Sarao R, Oudit GY, Yagil C, Kozieradzki I, Scanga SE, Oliveira-dos-Santos AJ, da Costa J, Zhang L, Pei Y, Scholey J, Ferrario CM, Manoukian AS, Chappell MC, Backx PH, Yagil Y, Penninger JM. Angiotensin-converting enzyme 2 is an essential regulator of heart function. *Nature* 2002; **417**: 822-828 [PMID: 12075344 DOI: 10.1038/nature00786]

57 **Gurley SB**, Allred A, Le TH, Griffiths R, Mao L, Philip N, Haystead TA, Donoghue M, Breitbart RE, Acton SL, Rockman HA, Coffman TM. Altered blood pressure responses and normal cardiac phenotype in ACE2-null mice. *J Clin Invest* 2006; **116**: 2218-2225 [PMID: 16878172 DOI: 10.1172/JCI16980]

58 **Chappell MC**. Does ACE2 contribute to the development of hypertension? *Hypertens Res* 2010; **33**: 107-109 [PMID: 20019704 DOI: 10.1038/hr.2009.207]

59 **Yamamoto K**, Ohishi M, Katsuya T, Ito N, Ikushima M, Kaibe M, Tatara Y, Shiota A, Sugano S, Takeda S, Rakugi H, Ogihara T. Deletion of angiotensin-converting enzyme 2 accelerates pressure overload-induced cardiac dysfunction by increasing local angiotensin II. *Hypertension* 2006; **47**: 718-726 [PMID: 16505206 DOI: 10.1161/01.HYP.0000205833.89478.5b]

60 **Tikellis C**, Cooper ME, Bialkowski K, Johnston CI, Burns WC, Lew RA, Smith AI, Thomas MC. Developmental expression of ACE2 in the SHR kidney: a role in hypertension? *Kidney Int* 2006; **70**: 34-41 [PMID: 16710353 DOI: 10.1038/sj.ki.5000428]

61 **Shaltout HA**, Figueroa JP, Rose JC, Diz DI, Chappell MC. Alterations in circulatory and renal angiotensin-converting enzyme and angiotensin-converting enzyme 2 in fetal programmed hypertension. *Hypertension* 2009; **53**: 404-408 [PMID: 19047579 DOI: 10.1161/HYPERTENSIONAHA.108.124339]

62 **Rentzsch B**, Todiras M, Iliescu R, Popova E, Campos LA, Oliveira ML, Baltatu OC, Santos RA, Bader M. Transgenic angiotensin-converting enzyme 2 overexpression in vessels of SHRSP rats reduces blood pressure and improves endothelial function. *Hypertension* 2008; **52**: 967-973 [PMID: 18809792 DOI: 10.1161/HYPERTENSIONAHA.108.114322]

63 **Lo J**, Patel VB, Wang Z, Levasseur J, Kaufman S, Penninger JM, Oudit GY. Angiotensin-converting enzyme 2 antagonizes angiotensin II-induced pressor response and NADPH oxidase activation in Wistar-Kyoto rats and spontaneously hypertensive rats. *Exp Physiol* 2013; **98**: 109-122 [PMID: 22750422 DOI: 10.1113/expphysiol.2012.067165]

64 **Ye M**, Wysocki J, Gonzalez-Pacheco FR, Salem M, Evora K, Garcia-Halpin L, Poglitsch M, Schuster M, Batlle D. Murine recombinant angiotensin-converting enzyme 2: effect on angiotensin II-dependent hypertension and distinctive angiotensin-converting enzyme 2 inhibitor characteristics on rodent and human angiotensin-converting enzyme 2. *Hypertension* 2012; **60**: 730-740 [PMID: 22777933 DOI: 10.1161/HYPERTENSIONAHA.112.198622]

65 **Patel SK**, Wai B, Ord M, MacIsaac RJ, Grant S, Velkoska E, Panagiotopoulos S, Jerums G, Srivastava PM, Burrell LM. Association of ACE2 genetic variants with blood pressure, left ventricular mass, and cardiac function in Caucasians with type 2 diabetes. *Am J Hypertens* 2012; **25**: 216-222 [PMID: 21993363 DOI: 10.1038/ajh.2011.188]

66 **Luo Y**, Liu C, Guan T, Li Y, Lai Y, Li F, Zhao H, Maimaiti T, Zeyaweiding A. Association of ACE2 genetic polymorphisms with hypertension-related target organ damages in south Xinjiang. *Hypertens Res* 2019; **42**: 681-689 [PMID: 30542083 DOI: 10.1038/s41440-018-0166-6]

67 **Fan X**, Wang Y, Sun K, Zhang W, Yang X, Wang S, Zhen Y, Wang J, Li W, Han Y, Liu T, Wang X, Chen J, Wu H, Hui R; Study Group for Pharmacogenomic Based Antihypertensive Drugs Selection, Effects and Side Effects, in Rural Area Chinese. Polymorphisms of ACE2 gene are associated with essential hypertension and antihypertensive effects of Captopril in women. *Clin Pharmacol Ther* 2007; **82**: 187-196 [PMID: 17473847 DOI: 10.1038/sj.clpt.6100214]

68 **Fan R**, Mao SQ, Gu TL, Zhong FD, Gong ML, Hao LM, Yin FY, Dong CZ, Zhang LN. Preliminary analysis of the association between methylation of the ACE2 promoter and essential hypertension. *Mol Med Rep* 2017; **15**: 3905-3911 [PMID: 28440441 DOI: 10.3892/mmr.2017.6460]

69 **Tucker NR**, Chaffin M, Bedi KC Jr, Papangeli I, Akkad AD, Arduini A, Hayat S, Eraslan G, Muus C, Bhattacharyya RP, Stegmann CM; Human Cell Atlas Lung Biological Network, Margulies KB, Ellinor PT; Human Cell Atlas Lung Biological Network Consortium Members. Myocyte-Specific Upregulation of *ACE2* in Cardiovascular Disease: Implications for SARS-CoV-2-Mediated Myocarditis. *Circulation* 2020; **142**: 708-710 [PMID: 32795091 DOI: 10.1161/CIRCULATIONAHA.120.047911]

70 **Ramchand J**, Burrell LM. Circulating ACE2: a novel biomarker of cardiovascular risk. *Lancet* 2020; **396**: 937-939 [PMID: 33010824 DOI: 10.1016/S0140-6736(20)32011-0]

71 **Huentelman MJ**, Grobe JL, Vazquez J, Stewart JM, Mecca AP, Katovich MJ, Ferrario CM, Raizada MK. Protection from angiotensin II-induced cardiac hypertrophy and fibrosis by systemic lentiviral delivery of ACE2 in rats. *Exp Physiol* 2005; **90**: 783-790 [PMID: 16049057 DOI: 10.1113/expphysiol.2005.031096]

72 **Díez-Freire C**, Vázquez J, Correa de Adjounian MF, Ferrari MF, Yuan L, Silver X, Torres R, Raizada MK. ACE2 gene transfer attenuates hypertension-linked pathophysiological changes in the SHR. *Physiol Genomics* 2006; **27**: 12-19 [PMID: 16788004 DOI: 10.1152/physiolgenomics.00312.2005]

73 **Zhang C**, Zhao YX, Zhang YH, Zhu L, Deng BP, Zhou ZL, Li SY, Lu XT, Song LL, Lei XM, Tang WB, Wang N, Pan CM, Song HD, Liu CX, Dong B, Zhang Y, Cao Y. Angiotensin-converting enzyme 2 attenuates atherosclerotic lesions by targeting vascular cells. *Proc Natl Acad Sci U S A* 2010; **107**: 15886-15891 [PMID: 20798044 DOI: 10.1073/pnas.1001253107]

74 **Úri K**, Fagyas M, Kertész A, Borbély A, Jenei C, Bene O, Csanádi Z, Paulus WJ, Édes I, Papp Z, Tóth A, Lizanecz E. Circulating ACE2 activity correlates with cardiovascular disease development. *J Renin Angiotensin Aldosterone Syst* 2016; **17** [PMID: 27965422 DOI: 10.1177/1470320316668435]

75 **Epelman S**, Shrestha K, Troughton RW, Francis GS, Sen S, Klein AL, Tang WH. Soluble angiotensin-converting enzyme 2 in human heart failure: relation with myocardial function and clinical outcomes. *J Card Fail* 2009; **15**: 565-571 [PMID: 19700132 DOI: 10.1016/j.cardfail.2009.01.014]

76 **Narula S**, Yusuf S, Chong M, Ramasundarahettige C, Rangarajan S, Bangdiwala SI, van Eikels M, Leineweber K, Wu A, Pigeyre M, Paré G. Plasma ACE2 and risk of death or cardiometabolic diseases: a case-cohort analysis. *Lancet* 2020; **396**: 968-976 [PMID: 33010842 DOI: 10.1016/S0140-6736(20)31964-4]

77 **Calò LA**, Schiavo S, Davis PA, Pagnin E, Mormino P, D'Angelo A, Pessina AC. ACE2 and angiotensin 1-7 are increased in a human model of cardiovascular hyporeactivity: pathophysiological implications. *J Nephrol* 2010; **23**: 472-477 [PMID: 20349406 DOI: 10.1089/end.2010.0121]

78 **Iwai M**, Horiuchi M. Devil and angel in the renin-angiotensin system: ACE-angiotensin II-AT1 receptor axis vs. ACE2-angiotensin-(1-7)-Mas receptor axis. *Hypertens Res* 2009; **32**: 533-536 [PMID: 19461648 DOI: 10.1038/hr.2009.74]

79 **Tan WSD**, Liao W, Zhou S, Mei D, Wong WF. Targeting the renin-angiotensin system as novel therapeutic strategy for pulmonary diseases. *Curr Opin Pharmacol* 2018; **40**: 9-17 [PMID: 29288933 DOI: 10.1016/j.coph.2017.12.002]

80 **Morrell NW**, Stenmark KR. The renin-angiotensin system in pulmonary hypertension. *Am J Respir Crit Care Med* 2013; **187**: 1138-1139 [PMID: 23675718 DOI: 10.1164/rccm.201210-1872LE]

81 **Zhang PX**, Han CH, Zhou FJ, Li L, Zhang HM, Liu WW. Renin-angiotensin system and its role in hyperoxic acute lung injury. *Undersea Hyperb Med* 2016; **43**: 239-246 [PMID: 27416692]

82 **Imai Y**, Kuba K, Rao S, Huan Y, Guo F, Guan B, Yang P, Sarao R, Wada T, Leong-Poi H, Crackower MA, Fukamizu A, Hui CC, Hein L, Uhlig S, Slutsky AS, Jiang C, Penninger JM. Angiotensin-converting enzyme 2 protects from severe acute lung failure. *Nature* 2005; **436**: 112-116 [PMID: 16001071 DOI: 10.1038/nature03712]

83 **Sturrock BR**, Milne KM, Chevassut TJ. The renin-angiotensin system - a therapeutic target in COVID-19? *Clin Med (Lond)* 2020; **20**: e72-e75 [PMID: 32414711 DOI: 10.7861/clinmed.2020-0146]

84 **Kuba K**, Imai Y, Rao S, Gao H, Guo F, Guan B, Huan Y, Yang P, Zhang Y, Deng W, Bao L, Zhang B, Liu G, Wang Z, Chappell M, Liu Y, Zheng D, Leibbrandt A, Wada T, Slutsky AS, Liu D, Qin C, Jiang C, Penninger JM. A crucial role of angiotensin converting enzyme 2 (ACE2) in SARS coronavirus-induced lung injury. *Nat Med* 2005; **11**: 875-879 [PMID: 16007097 DOI: 10.1038/nm1267]

85 **Mohamed TL**, Nguyen HT, Abdul-Hafez A, Dang VX, Dang MT, Gewolb IH, Uhal BD. Prior hypoxia prevents downregulation of ACE-2 by hyperoxia in fetal human lung fibroblasts. *Exp Lung Res* 2016; **42**: 121-130 [PMID: 27093376 DOI: 10.3109/01902148.2016.1157712]

86 **Koch A**, Kähler W, Klapa S, Grams B, van Ooij PAM. The conundrum of using hyperoxia in COVID-19 treatment strategies: may intermittent therapeutic hyperoxia play a helpful role in the expression of the surface receptors ACE2 and Furin in lung tissue via triggering of HIF-1α? *Intensive Care Med Exp* 2020; **8**: 53 [PMID: 32953400 DOI: 10.1186/s40635-020-00323-1]

87 **Zhang X**, Zheng J, Yan Y, Ruan Z, Su Y, Wang J, Huang H, Zhang Y, Wang W, Gao J, Chi Y, Lu X, Liu Z. Angiotensin-converting enzyme 2 regulates autophagy in acute lung injury through AMPK/mTOR signaling. *Arch Biochem Biophys* 2019; **672**: 108061 [PMID: 31356776 DOI: 10.1016/j.abb.2019.07.026]

88 **Coutard B**, Valle C, de Lamballerie X, Canard B, Seidah NG, Decroly E. The spike glycoprotein of the new coronavirus 2019-nCoV contains a furin-like cleavage site absent in CoV of the same clade. *Antiviral Res* 2020; **176**: 104742 [PMID: 32057769 DOI: 10.1016/j.antiviral.2020.104742]

89 **Li G**, He X, Zhang L, Ran Q, Wang J, Xiong A, Wu D, Chen F, Sun J, Chang C. Assessing ACE2 expression patterns in lung tissues in the pathogenesis of COVID-19. *J Autoimmun* 2020; **112**: 102463 [PMID: 32303424 DOI: 10.1016/j.jaut.2020.102463]

90 **Li Z**, Li X, Shen J, Tan H, Rong T, Lin Y, Feng E, Chen Z, Jiao Y, Liu G, Zhang L, Vai Chan MT, Kei Wu WK. Bioinformatic analysis of SMN1-ACE/ACE2 interactions hinted at a potential protective effect of spinal muscular atrophy against COVID-19-induced lung injury. *Brief Bioinform* 2021; **22**: 1291-1296 [PMID: 33190150 DOI: 10.1093/bib/bbaa285]

91 **Winkler ES**, Bailey AL, Kafai NM, Nair S, McCune BT, Yu J, Fox JM, Chen RE, Earnest JT, Keeler SP, Ritter JH, Kang LI, Dort S, Robichaud A, Head R, Holtzman MJ, Diamond MS. SARS-CoV-2 infection of human ACE2-transgenic mice causes severe lung inflammation and impaired function. *Nat Immunol* 2020; **21**: 1327-1335 [PMID: 32839612 DOI: 10.1038/s41590-020-0778-2]

92 **Han K**, Blair RV, Iwanaga N, Liu F, Russell-Lodrigue KE, Qin Z, Midkiff CC, Golden NA, Doyle-Meyers LA, Kabir ME, Chandler KE, Cutrera KL, Ren M, Monjure CJ, Lehmicke G, Fischer T, Beddingfield B, Wanek AG, Birnbaum A, Maness NJ, Roy CJ, Datta PK, Rappaport J, Kolls JK, Qin X. Lung Expression of Human Angiotensin-Converting Enzyme 2 Sensitizes the Mouse to SARS-CoV-2 Infection. *Am J Respir Cell Mol Biol* 2021; **64**: 79-88 [PMID: 32991819 DOI: 10.1165/rcmb.2020-0354OC]

93 **Tseng YH**, Yang RC, Lu TS. Two hits to the renin-angiotensin system may play a key role in severe COVID-19. *Kaohsiung J Med Sci* 2020; **36**: 389-392 [PMID: 32492292 DOI: 10.1002/kjm2.12237]

94 **Khan A**, Benthin C, Zeno B, Albertson TE, Boyd J, Christie JD, Hall R, Poirier G, Ronco JJ, Tidswell M, Hardes K, Powley WM, Wright TJ, Siederer SK, Fairman DA, Lipson DA, Bayliffe AI, Lazaar AL. A pilot clinical trial of recombinant human angiotensin-converting enzyme 2 in acute respiratory distress syndrome. *Crit Care* 2017; **21**: 234 [PMID: 28877748 DOI: 10.1186/s13054-017-1823-x]

**Footnotes**

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**Figure Legends**



**Figure 1 Composition and function of the renin-angiotensin system and its main regulators and inhibitors.** RAS: Rennin angiotensin system; ACEI: Angiotensin-converting enzyme inhibitor; ARB: Angiotensin II receptor blocker; ACE: Angiotensin-converting enzyme; ACE2: Angiotensin-converting enzyme 2; AT1R: Angiotensin type 1 receptor; AT2R: Angiotensin type 2 receptor; MasR: Mas receptor; MLN-4760: A specific ACE2 inhibitor; Organ damage: Renal damage, lung damage, cardiovascular damage, *etc.*

**Table 1 List of animal studies on the high-sodium diet and angiotensin-converting enzyme 2**

|  |  |  |  |
| --- | --- | --- | --- |
| **Animal** | **Study details** | **Main findings** | **Ref.** |
| Zucker rats | Lean and obese Zucker rats were fed a normal-sodium diet (0.4%) or a high-sodium diet (8%) for 2 wk | ACE2 mRNA and protein expression was significantly reduced in HS-fed obese Zucker rats | Samuel *et al*[19] |
| Wistar rats | Rats were fed three salt concentrations (0.2%, 1.2%, 8.2%) for 4 wk after uninephrectomy of the left kidney | HS diet increased the glomerular ACE/ACE2 ratio | Bernardi *et al*[20] |
| Wistar rats | After 4 wk of induction of hypertension (2-kidney 1-clip model), rats were fed a normal-sodium diet (0.4%) or a high-sodium diet (8%) for 2 wk | HS diet decreased urinary angiotensinogen, ACE, and ACE2 expression in the clipped and unclipped kidneys | Shimoura *et al*[21] |
| SHR | Rats were fed three NaCl content diets (0.03%, 0.3%, 3%) for 6 mo | HS diet decreased ACE2 protein expression in kidney tissues | Berger *et al*[18] |
| SHR | Rats were fed an 8% salt diet for 5 wk | HS diet decreased cardiac ACE2 mRNA and protein levels | Varagic *et al*[22] |
| DS and DR rats | DS and DR rats were fed low-sodium chow (0.45%) or high-sodium chow (7%) for 8 wk and treated with or without eplerenone (100 mg/kg/d), candesartan (10 mg/kg/d), or both drugs for 8 wk | HS diet increased angiotensinogen mRNA and decreased *ACE2* mRNA in the hearts of DS rats; candesartan increased *ACE2* mRNA levels in the heart | Takeda *et al*[23] |
| SD rats | Rats were fed an 8% NaCl high-salt or 0.4% NaCl (normal-salt) diet for 3 wk, with or without antioxidant supplementation with tempol | HS diet decreased ACE2 expression; tempol reversed the imbalance of renal RAS components (decrease in Ang II and AT1R and increase in AT2, ACE2, Ang 1-7, and MasR staining intensity) | Cao *et al*[24] |

SHR: Spontaneously hypertensive rats; DS rats: Dahl salt-sensitive hypertensive rats; DR rats: Dahl salt-resistant rats; SD rats: Sprague-Dawley rats; HS: High-sodium; ACE2: Angiotensin-converting enzyme 2; MasR: Mas receptor; AT1R: Angiotensin type 1 receptor; AT2: Angiotensin type 2; Ang 1-7: Angiotensin 1-7; RAS: Renin angiotensin system.

**Table 2 List of animal studies on renin angiotensin system inhibitors and angiotensin-converting enzyme 2**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Animal** | **Model** | **Study details** | **Main findings** | **Ref.** |
| Lewis rats | Normotensive | Rats were assigned to drink water containing losartan or lisinopril at 10 mg/kg/d for 12 d | Lisinopril or losartan increased cardiac *ACE2* mRNA, but the combination did not produce this effect | Ferrario *et al*[26] |
| SD rats | MI | Enalapril was given to rats after sham operation or LCA ligation | Enalapril prevented the decrease of ventricular *ACE2* mRNA levels and activities post-MI | Ocaranza *et al*[27] |
| Lewis rats | MI | Losartan and olmesartan was administered for 28 d after coronary artery ligation | The level of *ACE2* mRNA increased after treatment with losartan and olmesartan | Ishiyama *et al*[28] |
| C57BLKS/J mice | Normotensive | Mice were treated with telmisartan at 2 mg/kg/d for 2 wk | Telmisartan increased ACE2 expression in the tunica media of renal arterioles | Soler *et al*[29] |
| SD rats | MI | SD rats received ramipril at 1 mg/kg/d for 28 d after MI operation | Ramipril inhibited cardiac ACE but had no effect on cardiac *ACE2* mRNA after MI | Burrell *et al*[30] |
| SD rats | MI | Ramipril (1 mg/kg/d) and valsartan (10 mg/kg/d) were given for 28 d after MI operation | Cardiac ACE2 expression was not augmented after either treatment alone or in combination | Burchill *et al*[31] |

MI: Myocardial infarction; LCA: Left ventricular artery; SD rats: Sprague-Dawley rats; ACE2: Angiotensin-converting enzyme 2.

**Table 3 List of animal studies on the role of angiotensin-converting enzyme 2 in diabetes, hypertension, cardiovascular disease, and acute lung injury**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Disease** | **Animal** | **Study details** | **Main findings** | **Ref.** |
| Diabetes | db/db mice | Mice were randomly assigned to four treatment groups: (1) Control group fed normal chow; (2) Control group fed rosiglitazone diet; (3) db/db group fed normal chow; and (4) db/db group fed rosiglitazone diet | Protein expression of glomerular ACE2 was decreased in the kidneys of db/db mice, while tubular ACE2 and ADAM17 were increased. Rosiglitazone treatment of db/db mice normalized hyperglycemia, attenuated renal injury, and decreased urinary ACE2 and renal ADAM17 protein expression | Chodavarapu *et al*[45] |
| db/db mice | Mice were treated for 16 wk with a specific ACE2 inhibitor (MLN-4760) alone or combined with telmisartan. | ACE and ACE2 colocalized on the apical surface of the proximal tubules, whereas in glomeruli, ACE2 is present in podocytes and, to a lesser extent, in glomerular mesangial cells, whereas ACE is present only in endothelial cells. Telmisartan prevented the increase in UAE associated with the ACE2 inhibitor | Ye *et al*[46] |
| db/db mice | ACE and ACE 2 expression was measured in the kidney and heart | ACE2 protein in renal cortical tubules was increased, whereas ACE protein was decreased. In heart tissue, there were no significant differences between db/db and db/m mice in either ACE or ACE2 expression | Ye *et a l*[48] |
| STZ-induced diabetic SD rats | ACE2 and ACE gene and protein expression was measured in the kidney | *ACE2* and *ACE* mRNA levels were decreased in diabetic renal tubules by approximately 50% and were not influenced by ramipril | Tikellis *et al*[47] |
| STZ-induced diabetic Wistar rats | Diabetic Wistar rats were treated with DIZE | Treatment with DIZE restored ACE2 expression in glomeruli and increased the expression of AT2 receptors in whole kidney and isolated glomeruli | Goru *et al*[44] |
| Akita and Ace2-/- mice | Ace2-/- mice were crossed with Akita mice (Ins2WT/C96Y), and four groups of mice were studied: Ace2+/yIns2WT/WT, Ace2-/yIns2WT/WT, Ace2+/yIns2WT/C96Y, and Ace2-/y Ins2WT/C96Y. The Ace2+/yIns2WT/C96Y and Ace2-/y Ins2WT/C96Y mice were treated with the ARB (irbesartan) | Deletion of the *ACE2* gene was associated with accelerated kidney injury and reduced ACE2 expression in diabetic mice. Irbesartan reduced urinary albumin excretion rate in Ace2-/y Ins2WT/C96Y mice | Wong *et al*[49] |
| STZ-induced diabeticC57BL/6J mice and ACE2 knockout (KO) mice | Control and diabetic C57BL/6J and ACE2 KO mice, after 5 wk without treatment, were randomized to receive the ACE inhibitor perindopril. Wild-type mice were further randomized to receive the selective ACE2 inhibitor MLN-4760 | Induction of diabetes in wild-type mice was associated with a reduction in renal ACE2 expression and decreased Ang 1-7. In diabetic mice receiving MLN-4760 and in ACE2 KO mice, diabetes-associated albuminuria was enhanced | Tikellis *et al*[50] |
| Akita mice | Male diabetic Akita mice (Ins2 (WT/C96Y)) and control C57BL/6J mice (Ins2(WT/WT)) were injected daily with placebo or with rhACE2 (2 mg/kg) for 4 wk | Treatment with rhACE2 increased plasma ACE2 activity, normalized blood pressure, and reduced the urinary albumin excretion | Oudit *et al*[51] |
| STZ-induced diabetic Wistar rats | Diabetic Wistar rats were divided into 5 groups: No-treatment group, adenoviral (Ad)-ACE2 group, Ad-green fluorescent protein (GFP) group, ACEI group receiving benazepril and Ad-ACE2 + ACEI group | Rats in Ad-ACE2 group exhibited reduced SBP, urinary albumin excretion, creatinine clearance, glomeruli sclerosis index, and renal malondialdehyde level; downregulated transforming growth factor (TGF)-β1, vascular endothelial growth factor (VEGF), and collagen IV protein expression; and increased renal superoxide dismutase activity. Ad-ACE2 and ACEI had similar effects, whereas combined use of Ad-ACE2 and ACEI offered no additional benefits | Liu *et al*[52] |
| Hypertension | Salt-sensitive Sabra hypertensive rats, SHR, and SHRSP | ACE2 expression levels were determined in the kidneys | ACE2 levels were reduced in all of these hypertensive rat strains | Crackower *et al*[56] |
| ACE2-deficient mice | Ang II peptide was administered by i.v. infusion in wild-type and ACE2-deficient mice | Blood pressure measurements were substantially higher in the ACE2-deficient mice | Gurley *et al*[57] |
| SHR and Wistar Kyoto (WKY) rats | Expression of ACE2 was examined in the kidney from SHR and normotensive WKY rats | The tubular expression of ACE2 fell while glomerular expression of ACE2 was paradoxically increased in the SHR | Tikellis *et al*[60] |
| Sheep | Sheep were administered with betamethasone or vehicle at the 80th day of gestation and delivered at term | Antenatal steroid treatment resulted in the chronic alteration of ACE and ACE2 in the circulatory and tubular compartments of adolescent sheep, which may contribute to the higher blood pressure in this model of fetal programming–induced hypertension | Shaltout *et al*[61] |
| Transgenic rats | Transgenic rats were generated in an SHRSP genetic background expressing human ACE2 in vascular smooth muscle cells by the use of the *SM22* promoter (SHRSP-ACE2 model) | Mean arterial blood pressure was reduced in SHRSP-ACE2, and the vasoconstrictive response to intraarterial administration of angiotensin II was attenuated | Rentzsch *et al*[62] |
| SHR | Male WKY rats were randomized to receive either placebo or rhACE2 and were subsequently infused with Ang II | Treatment with rhACE2 partly corrected the hypertension, NADPH oxidase activation, and increased superoxide generation in the heart, kidney, and blood vessels | Lo *et al*[63] |
| Mice | ACE2 activity was measured in kidney cortex from mice that had received injection of MLN-4760 or DX600 | A marked increase in serum ACE2 activity. Mouse ACE2 abolished the hypertension induced by Ang II infusion. These effects were blocked by MLN-4760 but not by DX600 | Ye *et al*[64] |
| Cardiovascular disease | ACE2-deficient mice | ACE2 mutant mice were generated, and heart parameters were measured | Genetic inactivation of ACE2 using homologous recombination resulted in increased AngII peptide levels, upregulation of hypoxia genes in the heart, and severe cardiac dysfunction | Crackower *et al*[56] |
| ACE2-deficient mice | Ang II peptide was administered by i.v. infusion in WT and ACE2-deficient mice | No evidence for a role of ACE2 in the regulation of cardiac structure or function was found | Gurley *et al*[57] |
| SD rats | Lentiviral vector encoding mouse ACE2 (lenti-mACE2) or GFP was injected intracardially in Sprague–Dawley rats | ACE2 overexpression resulted in protective effects on AngII-induced cardiac hypertrophy and fibrosis | Huentelman *et al*[71] |
| SHR | Lentiviral vector encoding mouse ACE2 (lenti-mACE2) or GFP was injected intracardially in SHR and normotensive WKY rats | ACE2 overexpression exerted protective effects on high BP and cardiac pathophysiology induced by hypertension in the SHR | Díez-Freire *et al*[72] |
| Rabbits | 66 male New Zealand white rabbits were fed an atherogenic chow and were randomly divided into three groups: Treatment with a suspension of Ad-ACE2, treatment with a suspension of Ad-EGFP, and no treatment | ACE2 inhibited the development of early atherosclerotic lesions by suppressing the growth of VSMCs and improving endothelial function | Zhang *et al*[73] |
| Acute lung injury | ACE2 mutant mice | Acid aspiration-induced, sepsis-induced, and endotoxin-induced acute lung injury animal models were generated. Mice received intraperitoneal injections of rhACE2 protein | ACE2 and AT2 protected mice from severe acute lung injury. rhACE2 can protect mice from severe acute lung injury | Imai *et al*[82] |
| ACE2 knockout mice | Mice were intranasally inoculated with SARS-CoV virus | SARS-CoV receptor ACE2 had a protective role in acute lung failure | Kuba *et al*[84] |
| BALB/c mice | LPS-induced acute lung injury mice were treated with ACE2 activator resorcinolnaphthalein (RES) or ACE2 inhibitor MLN-4760 | ACE2 activation can reduce the severity of LPS-induced acute lung injury *via* the AMPK/mTOR pathway | Zhang *et al*[87] |
| c57BL/6J mice | Transgenic mice expressing the human ACE2 receptor driven by the cytokeratin-18 (K18) gene promoter (K18-hACE2) | Intranasal inoculation of SARS-CoV-2 in K18-hACE2 mice resulted in high levels of viral infection in lungs | Winkler *et al*[91] |
| c57BL/6J mice | Mice expressing hACE2 in the lung were transduced by oropharyngeal delivery of the recombinant human adenovirus type 5 that expresses hACE2 (Ad5-hACE2) | Mice were infected with SARS-CoV-2 and developed interstitial pneumonia associated with perivascular inflammation, accompanied by a higher viral load in the lungs | Han *et al*[92] |

UAE: Urinary albumin excretion; STZ: Streptozotocin; SD rats: Sprague-Dawley rats; SHR: Spontaneously hypertensive rats; SHRSP: Stroke-prone spontaneously hypertensive rats; SM22: Smooth muscle 22α; LPS: Lipopolysaccharide; SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2; ACE2: Angiotensin-converting enzyme 2.

**Table 4 List of epidemiological studies on the role of angiotensin-converting enzyme 2 in diabetes, hypertension, cardiovascular disease, and acute lung injury**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Disease** | **Source country** | **Patients** | **Main findings** | **Ref.** |
| Diabetes | Canada | Renal biopsies from 13 diabetic and 8 control patients | ACE2 mRNA and protein expression were significantly reduced in both the glomeruli and proximal tubules of the diabetic patients | Reich *et al*[53] |
| Japan | 66 nondiabetic and 8 diabetic patients with biopsy-proven renal diseases | *ACE2* mRNA expression was not significantly changed in the diabetic patients | Konoshita *et al*[54] |
| China | 275 Uygur T2D patients and 272 nondiabetic Uygur individuals | *ACE2* SNPs rs1978124, rs2048683, rs2074192, rs233575, rs4240157, rs4646156, rs4646188, and rs879922 were associated with T2D | Liu e*t al*[55] |
| Hypertension | Australia | 503 Caucasian subjects with type 2 diabetes | Genetic variation in *ACE2* was associated with hypertension and reduced systolic function in men, and hypertension and increased LV mass in women | Patel *et al*[65] |
| China | 275 Uygur T2D patients and 272 nondiabetic Uygur individuals | *ACE2* SNPs rs2048683, rs233575, rs4240157, rs4646156, rs4646188, and rs879922 were associated with increased SBP, while rs2074192, rs4646188, and rs879922 were associated elevated DBP | Liu *et al*[55] |
| China | 402 hypertensive patients and 233 normotensive individuals | *ACE2* variant rs2074192 was associated with EH, while rs4240157, rs4646155, and rs4830542 were associated with EH- and hypertension-related atrial fibrillation and left atrial remodeling | Luo *et al*[66] |
| China | 3408 untreated hypertensive patients | The T allele of *ACE2* rs2106809 was found to confer a 1.6-fold risk for hypertension in women | Fan *et al*[67] |
| China | 96 patients with EH and 96 healthy controls | Aberrant methylation of the *ACE2* promoter may be associated with EH risk | Fan *et al*[68] |
| Cardiovascular disease | United States | 11 individuals with dilated cardiomyopathy, 15 individuals with hypertrophic cardiomyopathy, and 16 controls with nonfailing hearts from the Penn Human Heart Tissue Biobank | ACE2 expression was downregulated in fibroblasts, pericytes, and vascular smooth muscle but upregulated in cardiomyocytes in dilated cardiomyopathy and hypertrophic cardiomyopathy | Fan *et al*[67] |
| Hungary | 45 healthy individuals, 239 hypertensiveindividuals,141 patients with heart failure (HF) and reduced ejection fraction (HFrEF), and 47 patients with HF and preserved ejection fraction (HFpEF) | ACE2 activity was further increased in HFrEF patients. Serum ACE2 activity was negatively correlated with left ventricular systolic function in HFrEF | Úri *et al*[74] |
| United States | 113 patients with chronic systolic heart failure | Elevated plasma soluble ACE2 (sACE2) activity was associated with greater severity of myocardial dysfunction and was an independent predictor of adverse clinical events | Epelman *et al*[75] |
| 14 countries across five continents | 10753 Prospective Urban Rural Epidemiology participants | Increased concentration of plasma ACE2 was associated with a higher risk of incident heart failure, myocardial infarction, stroke, and diabetes | Narula *et al*[76] |
| Italy | Healthy subjects (C) and EH and Bartter's/Gitelman's (BS/GS) patients | ACE2 was significantly elevated in BS/GS compared with either C or EH | Calò *et al*[77] |
| China | 275 Uygur T2D patients and 272 nondiabetic Uygur individuals | *ACE2* SNPs rs2074192 and rs879922 were associated with carotid arteriosclerosis stenosis and *ACE2* SNPs rs2048683, rs4240157, rs4646156, rs4646188, and rs879922 were linked to heavier left heart remodeling | Liu *et al*[55] |
| Acute lung injury | United States | 44 patients with acute respiratory distress syndrome (ARDS) | GSK2586881, a rhACE2, was well-tolerated in patients with ARDS, and has been found to reduce Ang II levels and increase Ang 1-7 levels | Khan *et al*[94] |

T2D: type 2 diabetes; EH: Essential hypertension; ACE2: Angiotensin-converting enzyme 2; ARDS: Acute respiratory distress syndrome; SNP: Single nucleotide polymorphism; HF: Heart failure; HFrEF: Heart failure and reduced ejection fraction; HFpEF: Heart failure and preserved ejection fraction; Ang 1-7: Angiotensin 1-7.