

Research Highlight

UDCA, a novel strategy for preventing SARS-CoV-2 infection via FXR-mediated ACE2 downregulation

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Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has resulted in the current outbreak of coronavirus disease 2019 (COVID-19) in humans, causing worldwide health issues [1]. The periodic and unexpected consequences have made SARS-CoV-2 a severe threat to human beings, forcing researchers to explore the underlying mechanisms of infection. Angiotensin converting enzyme 2 (ACE2), a cell surface protein, acts as a specific functional receptor for SARS-CoV-2, as well as the earlier SARS-CoV, leading to viral entry into host cells [2]. Because ACE2 is widely distributed in various organs, including the lung, small intestine, kidney, heart muscle, and so on, people infected by SARS-CoV-2 suffer a wide variety of symptoms [3]. The first and critical step for SARS-CoV-2 infection is the binding of its spike (S) protein to ACE2, leading to its attachment and entry into the host cells, which attracts much attention in the fight against COVID-19 [3]. Accordingly, different approaches for blocking viral-host interactions have been identified, either targeting the S protein of the virus or targeting the host ACE2 receptor that helps viral entry.

Convalescent plasma (CP), a passive immunization therapy, has always been used as an intervention in the prophylaxis and treatment of many infectious diseases. The efficacy of CP in treating COVID-19 is closely related to the concentration of neutralizing antibodies to the receptor binding domain (RBD) and N-terminal domain (NTD) of the S protein [4]. Alternatively, monoclonal antibodies (mAbs) are likely to be the most useful strategy at present and should be administered during the early stage of infection, which is identical to CP treatment [5]. However, all mAbs targeting the viral S-RBD show reduced efficacy due to rapid mutation, a common feature in RNA viruses [5]. Immune-evading SARS-CoV-2 variants have always been emerging with global mortality. Consequently, it is suggested that monitoring novel variants should be considered for the most effective mAb therapy. Notably, both high-titer CP and mAbs may be more efficient for immunocompromised patients with B-cell depletion. Nanobodies (Nbs) and miniproteins show potential advantages comparable to

some mAbs, as both of them can bind more tightly to the S-protein and efficiently block viral infectivity at low doses [6,7]. However, both of them have been evaluated in preclinical studies, and further investigations are needed before they can enter clinical trials.

It has been reported that administration of human recombinant soluble ACE2 (hsACE2) could competitively saturate the S-protein, thereby neutralizing the virus. Monteil *et al.* [8] demonstrated that hsACE2 significantly inhibits SARS-CoV-2 infectivity and directly neutralizes the virus in human blood vessels and kidney organoids, as shown in Figure 1. A phase 2 clinical trial of hsACE2 (APN01) with 178 participants reported that hsACE2 improved clinical outcome in patients treated with APN01 compared to placebo; however, the difference was not statistically significant. In addition to directly targeting the S-RBD or ACE2, interestingly, Razizadeh *et al.* [9] demonstrated that small molecules such as SSAA09E2 and nilotinib could interfere with and destabilize the ACE2-RBD complex with molecular dynamics (MD) analysis, which provides a new strategy to prevent infection at its early stages.

As mentioned above, CP, mAbs, Nbs, miniproteins and hsACE2 have relative limitations in preventing virus infection. Recently, Brevini *et al.* [10] demonstrated a novel strategy to protect against SARS-CoV-2 infection by reducing ACE2 expression. Due to the high expression of ACE2 and the production of infective viral progeny, gallbladder cholangiocyte organoids (GCOs) were used as a suitable study platform. They first identified chenodeoxycholic acid (CDCA) as a key agonist of ACE2 by activating transcription factor farnesoid X receptor (FXR) activity. Conversely, z-guggulsterone (ZGG) and ursodeoxycholic acid (UDCA), which are clinical drugs approved by the FDA, suppressed FXR activity and subsequently downregulated ACE2 levels, indicating that FXR directly regulates ACE2 expression in GCOs [10]. Considering the wide expression of FXR [11], they further revealed that UDCA or ZGG reduced ACE2 expression in primary airway and intestinal organoids via suppression of FXR, suggesting that FXR-mediated ACE2 regulation may represent a basic and general mechanism. Importantly, clinical doses of UDCA

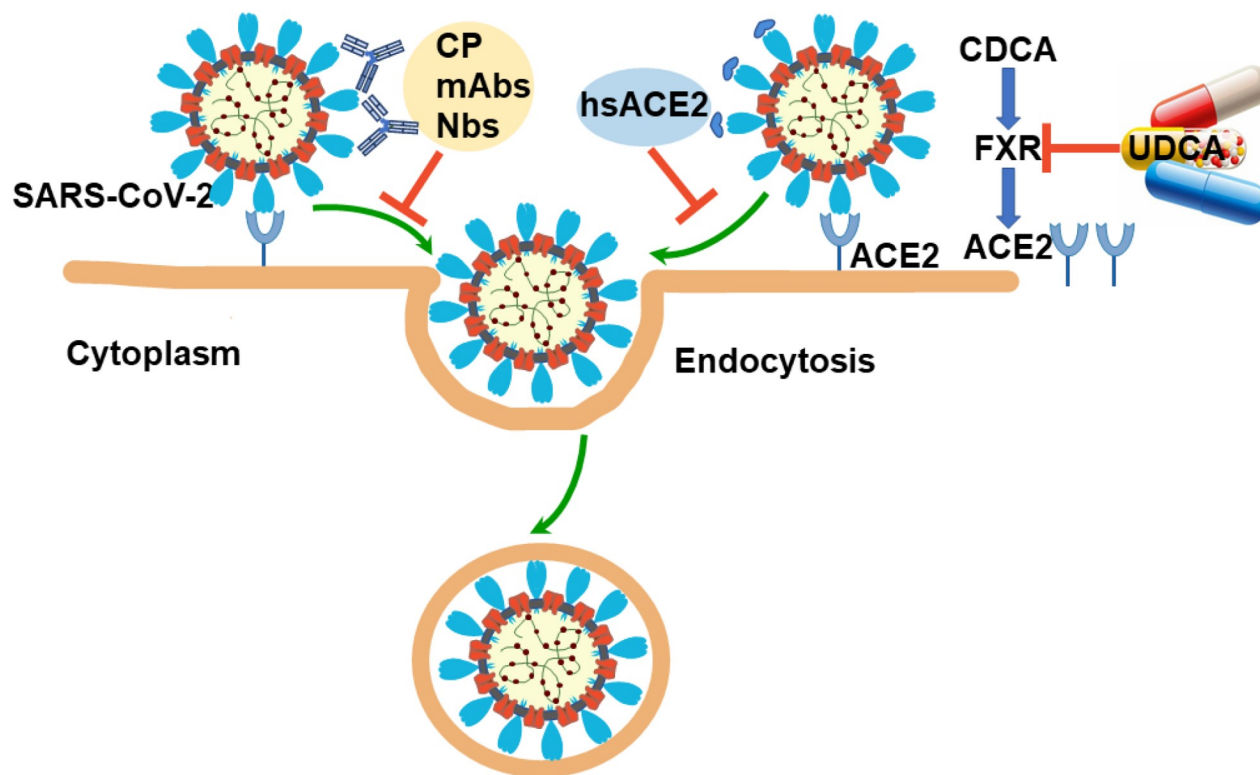


Figure 1. Inhibiting the binding of SARS-CoV-2 to ACE2 receptor decreases viral attachment and entry into the cells. Antibodies to the S-protein of SARS-CoV-2 inhibit virus attachment and binding to ACE2 receptors, thus inhibiting viral entry into the cell. UDCA inhibits ACE2 expression by inhibiting FXR activity, thereby limiting viral infection. CP: convalescent plasma; mAbs: monoclonal antibody; Nbs: nanobodies; CDCA: chenodeoxycholic acid; FXR: farnesoid X receptor; UDCA: ursodeoxycholic acid; ACE2: angiotensin-Converting Enzyme-2.

directly reduced SARS-CoV-2 infection *in vitro* (primary airway, biliary and intestinal organoids), *in vivo* (mice and hamsters), and in human lungs and livers *ex vivo* via FXR-mediated ACE2 downregulation, confirming the prophylactic potential of UDCA against COVID-19 [10], as shown in Figure 1.

All these encouraging results inspired them to evaluate the potential effect of UDCA on viral infection in humans. As expected, they illustrated that UDCA reduced ACE2 expression in the nasal epithelium in healthy volunteers. Based on these exciting findings, they finally identified a potential correlation between UDCA treatment and better clinical outcomes of COVID-19 patients, which was confirmed in two independent cohorts of liver transplant recipients and cirrhosis participants [10].

This work provides novel insight into reducing SARS-CoV-2 infection, which is in line with the conclusion of Mendelian randomization analyses, identifying ACE2 as a preferential therapeutic target for COVID-19 [12]. Second, ACE2 is a reasonable target, since it is a common receptor for other coronaviruses, including SARS-CoV, NeoCoV, and CoV-NL63, rendering it essential for viral entry [13]. Third, as ACE2 is a host protein with evolutionary conservation, therapies regulating ACE2 expression are unlikely to be affected by mutations of SARS-CoV-2. Last but not most importantly, UDCA has already been administered to patients in the clinic with good safety and minimal side effects [14] and showed no cytotoxic effects in the study. Taken together, UDCA, as an advantageous ACE2 modulator, warrants further consideration in large cohort studies to evaluate its role in SARS-CoV-2 infection.

Along with the aforementioned strategy, some points should be taken into consideration. For instance, FXR activation has been demonstrated to inhibit inflammation [11], while UDCA has also been shown to reduce inflammation in livers independent of FXR activity [15]. Thus, the multifaceted correlation between FXR activity, UDCA, viral infection and inflammation, as well as how to balance SARS-CoV-2 infection and inflammation, should be carefully considered. In addition, FXR is a target in the prevention of COVID-19, and whether there is a better FXR inhibitor that is more efficient for limiting viral infection remains unknown. More importantly, ACE2, as a primary regulator of both the renin-angiotensin-aldosterone system (RAAS) and the kinin-kallikrein system (KKS), plays a critical role in regulating blood pressure, fibrosis, coagulation and inflammation [16]. Thus, UDCA-mediated decreases in ACE2 expression may have paradoxical impacts, restricting SARS-CoV-2 infection and causing viral pathogenicity via disruption of the RAAS and KKS balance, particularly in patients with obesity, hypertension, diabetes mellitus, chronic pulmonary disease and cardiovascular diseases.

Taken together, a deep understanding of the role that ACE2 plays in various signaling pathways to maintain organismal physiology may help develop effective drugs for COVID-19.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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