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Potential soluble angiotensin-converting enzyme 2 in oral and salivary coronavirus infection therapy

Abstract

A comprehensive search was carried out in mainstream bibliographic databases or Medical Subject Headings, including ScienceDirect, PubMed, Scopus, and ISI Web of Science. The search was applied to the articles that were published between 2004 and early 2022. With strict literature search and screening processes, it yielded 4 articles from 142 articles of initial literature database. In oral cavity, tongue has the highest angiotensin-converting enzyme 2 (ACE 2) expression and lesser amounts in the other oral tissues, oral mucosa, including the gingival tissue. By Pre-incubation with SARS-CoV-2 (COVID-19) RBD, CTB-ACE 2 activity was absolutely inhibited, offering an the description for decreased saliva ACE 2 activity in COVID-19 patients. Through minimizing or debulking virus transmission, SARS-CoV-2 (COVID-19)-trapping proteins proposes an affordable strategy for protecting people from most oral re-infection, whereas newly evolving strains have higher viral load in saliva and greater transmission. Delta variant viral load in a patient is about 1,260 times higher than those infected with previous strains.

In conclusion, ACE 2 fusion proteins or chewing gum can be used as the rapid methods of decreasing SARS-CoV-2 (COVID-19) from saliva and oral cavity of the infected patients for minimizing infection and transmission, diagnosis, inhibitors, vaccine development, and therapy of SARS-CoV-2 (COVID-19) disease.

Keywords: ACE 2, angiotensin-converting enzyme 2, chewing gum, COVID-19, oral, salivary, SARS-CoV-2, soluble

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Abbreviations: ACE 2, angiotensin-converting enzyme 2; AKI, acute kidney injury; Ang II, angiotensin II; AT 1,2, alveolar epithelial cells type 2; CKD, chronic kidney disease; COVID-19, coronavirus disease-2019; CTB, cholera toxin B; DKD, diabetic kidney disease; IC_{50}, Ig, Immunoglobulin; Inhibitory Concentration at 50%, IgG, Immunoglobulin; mACE 2-Ig, fusion protein containing the ACE 2 variants; RAS, renin-angiotensin system; RBD, Receptor-Binding Domain; SARS-CoV-2, Severe-Acute-Respiratory- Syndrome Coronavirus type 2; TMPRSS 2, transmembrane protease serine 2

Objectives of the study

To seek a comfortably novel method of SARS-CoV-2 (COVID-19) salivary and oral infection treatment that can prevent the disease progression.

Introduction

In oral cavity, tongue has the highest angiotensin-converting enzyme 2 (ACE 2) expression and lesser amounts in the other oral tissues, oral mucosa, including the gingival tissue, in addition to other tissues of the human body, such as pulmonary alveolar epithelial cells type II (AT 2) or pneumocyte type II, myocardial cells (cardiomyocytes, expressed in myofibroblast and fibroblast in the stromal area spongiosa layer of the aortic valves), brush border of proximal renal tubular cells, urinary bladder urothelial cells, cholangiocytes (bile duct epithelial cells), ileum and colon enterocytes, and upper esophagus stratified epithelium. Higher mean ACE 2 expression was identified in the minor salivary glands, in comparison to the lungs.[1] The SARS-CoV and SARS-CoV-2 spike protein 1 bind to ACE 2 that located on the host cytomembrane through transmembrane protease serine 2 (TMPRSS 2), a cytomembrane protease.[2] Two previous histological studies in Chinese rhesus macaques and rats have revealed the ACE 2 presence in salivary glands.[3,4] Nevertheless, clearly unexplored distribution details of the ACE 2 and TMPRSS 2 in human saliva, salivary gland, oral and nasal epithelium remain.[5-7] A recent study demonstrated that *in vitro*, exogenous ACE 2 and TMPRSS 2 can anchor and fuse to oral mucosa and SARS-CoV-2 spike protein can bind to ACE 2 in human salivary glands.[5] Several previous studies revealed that there was expression of ACE 2 and TMPRSS 2 in human salivary glands.[8-11] The majority of the previous studies on the clinical manifestations have not verified the COVID-19 patients' oral health status.[12] The alteration in the taste perception could be considered an early manifestation in COVID-19 patients although the mechanism for taste alteration is not clearly defined.[13] A previous study in American cohort of 305 COVID-19 patients, aged from 4 to 60 years revealed that the ACE 2 expression was lower among patients aged between 4 and 9 years, compared with adolescents and adults,[14] while no cases of COVID-19 children with taste alteration have been reported,[13] including the lower risk of pulmonary injury and inflammation among the younger COVID-19 patients.[15,16] In addition to SARS-CoV-2 (COVID-19) transmission via direct inhalation of microdroplets spread by coughing, speaking, sneezing, shouting, and singing, direct contact with virus contaminated surfaces by self-dissemination via oral, nasal, and ophthalmic mucosa.[17] A recent study on oral health among COVID-19 patients during hospitalization demonstrated that 25 % of cases had taste impairment, 20 % of cases had swallowing difficulty, and 15 % of cases reported burning sensation of the tongue and oral cavity (present only in female patients).[18] A previous study in rodents revealed that gender- and age-dependent pattern of ACE 2 expression, with a more rapid decline with age in males, in comparison to females.[19] Additionally, 25 % of patients presented thyroid disorders (hyperthyroidism or hypothyroidism), 15 % of patients presented diabetes, and 15 % of patients presented obesity. There were no statistically significant results on the onset of some manifestations emerged between age and sex in this study.[18]

Method of the study

Search strategy and inclusion criteria

A comprehensive search was carried out in mainstream bibliographic databases or Medical Subject Headings, including ScienDirect, PubMed, Scopus, and ISI Web of Science. The search was applied to the articles that were published between 2004 and early 2022. Our first involved performing searches of article abstract/keywords/title using strings of ("SARS-CoV-2" or "COVID-19", "Angiotensin-Converting Enzyme 2" or "ACE 2", "Soluble", " Therapy", "Salivary" or "Saliva", "Oral"). After a first approach of search, published articles focusing on soluble ACE 2 therapy on oral and salivary COVID-19 infection were retained and the information on soluble ACE 2 in chewing gum and salivary and oral COVID-19 infection was extracted for having a crude knowledge involving their themes. Another round of publication search was conducted for adding the missing published articles that were not identified by the first round.

All keywords combinations from ACE 2, angiotensin-converting enzyme 2, chewing gum, COVID-19, oral, salivary, soluble, and SARS-CoV-2, to bind the population of cases were under consideration. Search string for disease groups include . "SARS-Cov-2" or "COVID-19" or "Oral" or "Salivary" or "ACE 2" or "Soluble ACE 2" or "Soluble Angiotensin-Converting Enzyme 2" or "Chewing" or "Gum ". The initial literature databases were further manually screened with the following rules: 1) non-oral or salivary COVID-19-related articles were excluded; 2) articles that did not report an oral or salivary COVID-19 treatment outcome related to (soluble) ACE 2 were not considered, such as commentary articles, or editorial; 3) non-peer reviewed articles were not considered to be of a scholarly trustworthy validity; and 4) duplicated and non-English articles were removed. The *in vitro*, *in vivo*, and clinical studied articles were carefully selected to guarantee the literature quality, which is a trade-off for quantity.

With strict literature search and screening processes, it yielded 4 articles from 142 articles of initial literature database. Needed article information was extracted from each article by: 1) direct information including journal, title, authors, abstract, full text documents of candidate studies, publishing year; 2) study period; 3) research method used; and 4) the conclusions made about the impact of soluble ACE 2 therapy on COVID-19-related oral and -related organ manifestations.

Results

From initial literature database of 142 articles, there were 4 published articles involving the ACE 2 therapy on COVID-19 as demonstrated in Table 1.

Table 1 Demonstrating published articles related to ACE 2 therapy on COVID-19

Published Year	Author (s)	Type of Study	Results/Conclusion	Reference
2019	Wysocki *et al*	*In Vivo*	Two short ACE 2 protein variants were systemically very active in mouse urine. Longer exposure of the 110 kDa native mouse recombinant ACE 2 to proteases from renal apical tubular membrane resulted in high ACE 2 enzymatic activity. Two active short ACE 2 proteins (1-619, 1-605) were around 4-5-fold higher than that of the native mouse recombinant ACE 2 protein. These short ACE 2 proteins were re-absorbable by the renal tubules, thus they could amplify renal ACE 2 protein activity and make attractive to fight against several renal diseases where the renin-angiotensin system (RAS) is over-active, such as acute-kidney- injury-associated COVID-19.	20
2020	Wrapp *et al*	*In Vivo*	SARS-CoV-2 S protein bound ACE 2 protein with higher affinity than did SARS-CoV S protein. SASRS-CoV Receptor-Binding Domain (RBD)-specific monoclonal antibodies did not have adequate binding to SARS-CoV-2, indicating that antibody-cross-reactivity between the two RBDs may be limited.	21
2020	Lei *et al*	*In Vitro*	Both fusion proteins (extracellular domain fusion recombinant human ACE 2 connecting to the Fc portion of the human immunoglobulin (IgG) 1, fusion mutant ACE 2 protein) demonstrated pharmacological properties in mice.	22
2022	Daniell *et al*	Case-Control	CTB-ACE 2 activity was completely inhibited by pre-incubation with SARS-CoV-2 RBD, providing the COVID-19 patients' reduction of the saliva ACE 2 activity. SARS-CoV-2-trapping- protein chewing gum demonstrated protecting COVID-19 patients from oral re-infections via debulking or virus transmission minimization.	23

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Discussion

ACE 2 protein is normally found in the urine. In mice model, small recombimant ACE 2 variants effectively degrade the excess of systemic circulating Angiotensin II (Ang II) and are both filtered and reabsorbed by the proximal renal tubules, therefore they can increase urinary ACE 2 activity and enhance blood pressure recovery.[20] Wrapp *et al* found that ACE 2 protein bound to SARS-CoV-2 (COVID-19) S ectodomain with approximately 15 nM affinity, that is approximately 10- to 20-fold higher than ACE 2 protein and formed complex of ACE 2 protein binding to SARS-CoV.[21] Increased RAS component activity in the kidney and urines was demonstrated both in rodent models of diabetic kidney disease (DKD) and in patients with DKD and non-diabetic CKD that contributing to both experimental and clinical acute kidney injury (AKI), both hemodynamic and non-hemodynamic mechanisms.[20] Lei et al. showed that ACE 2 fusion proteins bund the RBDs of both SARS-CoV-2 (COVID-19) and SARS-CoV with a high affinity contributing to potently neutralized by ACE-Ig and mACE 2-Ig.[21] The IC_{50} values of ACE 2-Ig for SARS-CoV-2 (COVID-19) and SARS-CoV were 0.1 µg/mL and 0.8 µg/mL, and the IC_{50} values of mACE 2-Ig for the neutralization of these two viruses were 0.08 µg/mL and 0.9 µg/mL, respectively. Most of the current human antibodies with potent neutralizing activity to the SARS-CoV reveal no cross-reactivity to SARS-CoV-2. Based on the pseudovirus system, neutralization of SARS-CoV-2 (COVID-19) with ACE 2-Ig can be targeted.[22] Recently, Daniell et al. prepared ACE 2 chewing gum tablet containing ground plant powder (expressed CTB-ACE 2 up to 17.2 mg ACE 2/g dry weight (11.7 % leaf protein, having physical characteristics and taste/flavor like conventional gums) by a compression process without protein loss during gum compression) for debulking and bocking of SARS-CoV-2 (COVID-19) entry into human cells and demonstrated that ACE 2 activity was markedly decreased in 10 saliva samples of the COVID-19 patients compared with control group (2,582 +/- 439.82 versus 50,126 +/- 2,101, change in relative fluorescence units : 27.63 +/- 9.52 versus 225 +/- 30.82 mU/mg enzyme activity units). All 10 COVID-19 saliva specimens revealed a similar and almost undetectable ACE 2 activity, but one specimen demonstrated as an outlier with enzyme activity (38,504 +/- 9,688 relative fluorescence units; 236.4 +/- 60.28 mU/mg enzyme activity units) similar to healthy saliva. This patient was diagnosed of asymptomatic SARS-CoV-2 (COVID-19)-saliva-positive PCR. By Pre-incubation with SARS-CoV-2 (COVID-19) RBD, CTB-ACE 2 activity was absolutely inhibited, offering an the description for decreased saliva ACE 2 activity in COVID-19 patients. Through minimizing or debulking virus transmission, SARS-CoV-2 (COVID-19)-trapping proteins proposes an affordable strategy for protecting people from most oral re-infection, whereas newly evolving strains have higher viral load in saliva and greater transmission. Delta variant viral load in a patient is about 1,260 times higher than those infected with previous strains.[23]

Conclusion

ACE 2 fusion proteins or chewing gum can be used as the rapid methods of decreasing SARS-CoV-2 (COVID-19) from saliva and oral cavity of the infected patients for minimizing infection and transmission, diagnosis, inhibitors, vaccine development, and therapy of SARS-CoV-2 (COVID-19) disease.

Authors contributions

Dr. Attapon Cheepsattayakorn conducted the study framework and wrote the manuscript. Associate Professor Dr. Ruangrong Cheepsattayakorn and Professor Dr. Porntep Siriwanarangsun contributed to scientific content and assistance in manuscript writing. All authors read and approved the final version of the manuscript.

Competing interests

The authors declare that they have no actual or potential competing financial interests.

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References

1. Xu H, Zhong L, Deng J, et al. High expression of ACE 2 receptor of 2019-nCoV on the epithelial cells of oral mucosa. *Int J Oral Sci*. 2020;12 (1):1–5.
2. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE 2 and TMPRSS 2 and is blocked by a clinically proven protease inhibitor. *Cell.* 2020;181(2): 271–280.
3. Cano IP, Dionisio TJ, Cestari TM, et al. Losartan and isoproterenol promote alterations in the local renin-angiotensin system of rat salivary glands. *PLoS.* 2019;14(5) :e0217030.
4. Liu L, Wei Q, Alvarez X, et al. Epithelial cells lining salivary gland ducts are early target cells of severe acute respiratory syndrome coronavirus infection in the upper respiratory tracts of rhesus macaques. *J Virol.* 2011;85(8):4025–4030.
5. Zhu F, Zhong Y, Ji H, et al. ACE 2 and TMPRSS 2 in human saliva can adsorb to the oral mucosal epithelium. *J Anat.* 2022; 240(2):398–409.
6. Hamming I, Timens W, Bulthuis ML,ert al. Tissue distribution of ACE 2 protein, the functional receptor for SARS coronavirus, a first step in understanding SARS pathogenesis. *J Pathol.* 2004;203(2):631–637.
7. Bertram S, Heurich A, Lavender H, et al. Influenza and SARS-coronavirus activating proteases TMPRSS 2 and HAT are expressed at multiple sites in human respiratory and gastrointestinal tracts. *PLoS One.* 2012;7 :e35876.
8. Pascolo L, Zupin L, Melato M, et al. TMPRSS 2 and ACE 2 co-expression in SARS-CoV-2 salivary gland infection. *J Dent Res.* 2020;99(10):1120–1121.
9. Shamsoddin E. Saliva: a diagnostic option and a transmission route for 2019-nCoV. *Evid Based Dent*. 2020;21(2):68–70.
10. Song J, Li Y, Huang X, et al. Systematic analysis of ACE 2 and TMPRSS 2 expression in salivary glands reveals underlying transmission mechanism caused by SARS-CoV-2. *J Med Virol*. 2020; 92 (11): 2556–2566.
11. Wang C, Wu H, Ding XU, et al. Does infection of 2019 novel coronavirus cause acute and/or chronic sialadenitis? *Med Hypotheses.* 2020; 140:109789.
12. Epidemiology Working Group for NCIP Epidemic Response, Chinese Center for Disease Control and Prevention.. The epidemiological characteristics of an outbreak of 2019 novel coronavirus diseases (COVID-19) in China. *Zhonghua Liu Xing Bing Xue Za Zhi.* 2020;41(2) :145–151.
13. Parra-Ortega I, Rodriguez-Ortega D. SARS-CoV-2 impact on oral health: a general view. *Bol Med Hosp Infant Mex*. 2021;78 (2): 91–94.
14. Patel A, Verma A. Nasal ACE 2 levels and COVID-19 in children. *JAMA.* 2020;323(23) :2386–2387.
15. Schouten L, Helmerhorst H, Wagenaar G, et al. Age-dependent changes in the pulmonary renin-angiotensin system are associated with severity of lung injury in a model of acute lung injury in rats. *Crit Care Med.* 2016;44(12) :e1226–e1235.
16. Bunyavanich S, Do A, Vicencio A. Nasal gene expression of angiotensin-converting enzyme 2 in children and adults. *JAMA*. 2020; 323(23) :2427–2429.

17. Xu R, Cui B, Duan X, et al. Saliva: potential diagnostic value and transmission of 2019-nCoV. *Int J oral Sci*. 2020; 12 :11.

18. Sinjari B, D´Ardes D, Santilli M, et al. SARS-CoV-2 and oral manifestation: an observational, human study. *J Clin Med.* 2020; 9(10):3218.

19. Xie X, Chen J, Wang X, et al. Age- and gender-related difference of ACE 2 expression in rat lung. *Life Sci.* 2006;78 (19):2166–2171.

20. Wysocki J, Schulze A, Batlle D. Novel variants of angiotensin-converting enzyme-2 of shorter molecular size to target the kidney renin-angiotensin system. *Biomolecules*. 2019;9(12):886.

21. Wrapp D, Wang N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion confirmation. *Science.* 2020; 367(6483):1260–1263.

22. Lei C, Qian K, Li T, et al. Neutralization of SARS-CoV-2 spike pseudotyped virus by recombinant ACE 2-Ig. *Nat Commun*. 2020; 11(1): 2070.

23. Daniell H, Nair SK, Esmaeili N, et al. Debulking SARS-CoV-2 in saliva using angiotensin-converting enzyme 2 in chewing gum to decrease oral virus transmission and infection. *Molecular Therapy*. 2022; 30 (4):13.

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