 doi:10.20944/preprints202004.0282.v1

Structural deviation and identification of novel inhibitor of SARS-CoV-2 spike protein through molecular docking; an *insilico* study

Anindya Sundar Panja*[1] and Aniket Sarkar[1]

1. Post-Graduate Department of Biotechnology, Molecular informatics Laboratory, Oriental Institute of Science and Technology, Vidyasagar University, Midnapore, West Bengal, India.

*To whom correspondence should be addressed: Anindya S. Panja: Department of Biotechnology, Molecular informatics Laboratory, Oriental Institute of Science and Technology, Vidyasagar University, Midnapore, West Bengal, India-721102; biotech2ani@gmail.com ; Tel.(+91)89007-89003. ORCID: - 0000-0003-2823-9948

Abstract

Purpose: Pandemic Novel Coronavirus (SARS-CoV-2) has emerger centered from wuhan, China. Structurally homologous spike protein of SARS-CoV-2 receptor is taxonomically homologous with SARS-Cov and SARS associated bat coronavirus. Still now scientists are trying to find out proper vaccine and treatments for this disease.

Methods: Systematically we modeled and compared the structure of SARS-CoV-2 spike protein along with Bat Cov, Bat SARS Cov and SARS Cov Urbani. S1 and S2 unit of the coronavirus (SARS-CoV-2) are attached with ACE2 and furin, here we docked 5 Ca+ chelating drugs with these two proteins.

Results: Structural comparison with all these spike proteins revealed that less significant but not negligible difference exists among them. Inserted stable nucleotide sequences and corresponding surface exposed peptidal region may be considered as epitope. Docking result with Toxicokinetics and half life of Penicillamine can effectly inhibit the attachment site of spike protein of coronavirus (SARS-CoV-2).

Conclusions: Docking summery and the pharmacokinetics with toxicokinetics index recommend that Penicillamine can able to inhibit the infection of SARS-CoV-2.

Keywords: SARS-CoV-2; Penicillamine; spike protein; Docking

Introduction

The recent pandemic novel coronavirus (2019-nCoV), caused by severe acute respiratory syndrome corona virus2 (SARS-CoV-2; first identified on December 12th, 2019) was initially detected in Wuhan, Hubei Province, China [1]. Coronaviruses infect many mammalian and avian species cause severe acute respiratory syndrome coronavirus (SARS-CoV) [2,3]. Coronaviruses having a single-stranded enveloped RNA viruses and the envelope-anchored spike protein with S1 and S2 units [4,5] recognize the host receptor angiotensin-converting enzyme 2 (ACE2) and polybasic cleavage site furin. By regulating furin-mediated substrate cleavage results a crucial role in pathogen infection, neurologic disease and cancer, already several drugs are already explore based on furin cleavage-targeted mechanism[6,7]. The genome of SARS-CoV-2 has been sequenced (Lu R et al., 2020) and it was reported that the structure of spike proteins of SARS-CoV-2 and SARS-CoV have a high degree of homology [8]. In our study, we perform *in-silico* sequence analyses and model structural homology of corona virus (2019-nCoV) and related four SARS-CoV viruses and calculate RMSD. As furin is calcium-dependent proteases, we have retrieved five selected calcium chelating drug from published literature based upon their therapeutic ability which may control the structural alteration in the spike protein that allows the viral entry process into host cells [9,10,11,12]. The selected drugs have been evaluated against ACE2 and Furin both by molecular docking. Based on our observation, we hypothesize the strategies for pharmacological intervention by targeting ACE2 and furin to combat corona virus (2019-nCoV) infection in human.

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 16 April 2020 doi:10.20944/preprints202004.0282.v1

Method

RNA and protein sequence ; protein structure modelling
RNA and amino acid sequences of the SARS-CoV-2 were downloaded from NCBI GenBank. Protein structure of the spike Glyco-proteins were modelled by Swiss Model Server (www.swissmodel.expa sy.org) and finally validated by Procheck.
Structural homology
PyMOL was used to calculate based on RMSD algorithm to estimate the structural deviation of spike glyco-proteins from different SARS-CoV viruses [13].
Molecular docking
ACE2 and Furin were taken as target and ligands (succimer, dimercaprol (BAL), edetate calcium disodium, deferoxamine, and penicillamine) were docked by using AutoDock 1.5.6 [14] (Supplementary 1 Table A-J).

Result

Insertion of new RNA sequence was already identified by Andersen KG., et al and he reported that 12 number of newly evolved nucleotides adopted by SARS-CoV-2 [15].

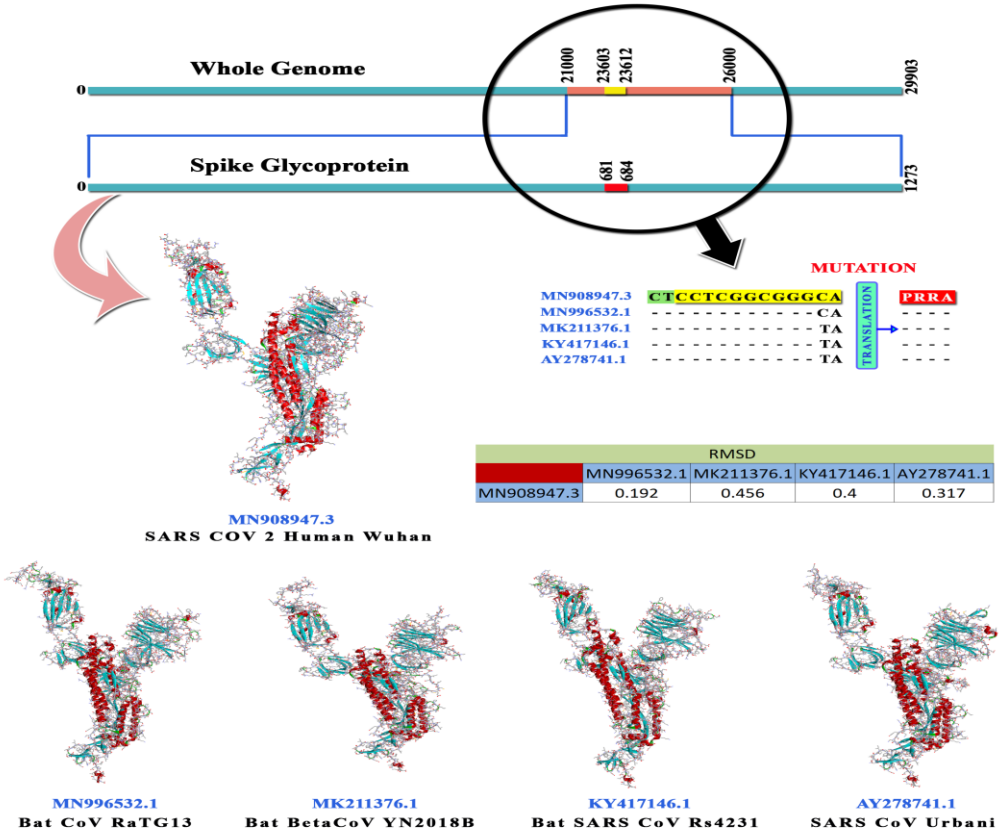

Figure 1. Sequence comparison and structural deviation of the spike glyco-protein of SARS-CoV-2 binds. Newly inserted Nucleotides highlighted as yellow and green colour, the corresponding amino acids are in Red colour. The structures of spike glyco-proteins are modelled and structural deviation (RMSD) are presented.

This newly inserted nucleotide sequences which is highly GC rich (CTCCTCGGCGGGCA) and the corresponding amino acids (PRRA), expressed on the surface of spike protein of SARS-CoV-2. The exposed peptidal region of the spike protein may help to screen epitope for the development of new vaccine to combat SARS-CoV-2 infection (Fig 1).

Preprints (www.preprints.org) | NOT PEER-REVIEWED | Posted: 16 April 2020 doi:10.20944/preprints202004.0282.v1

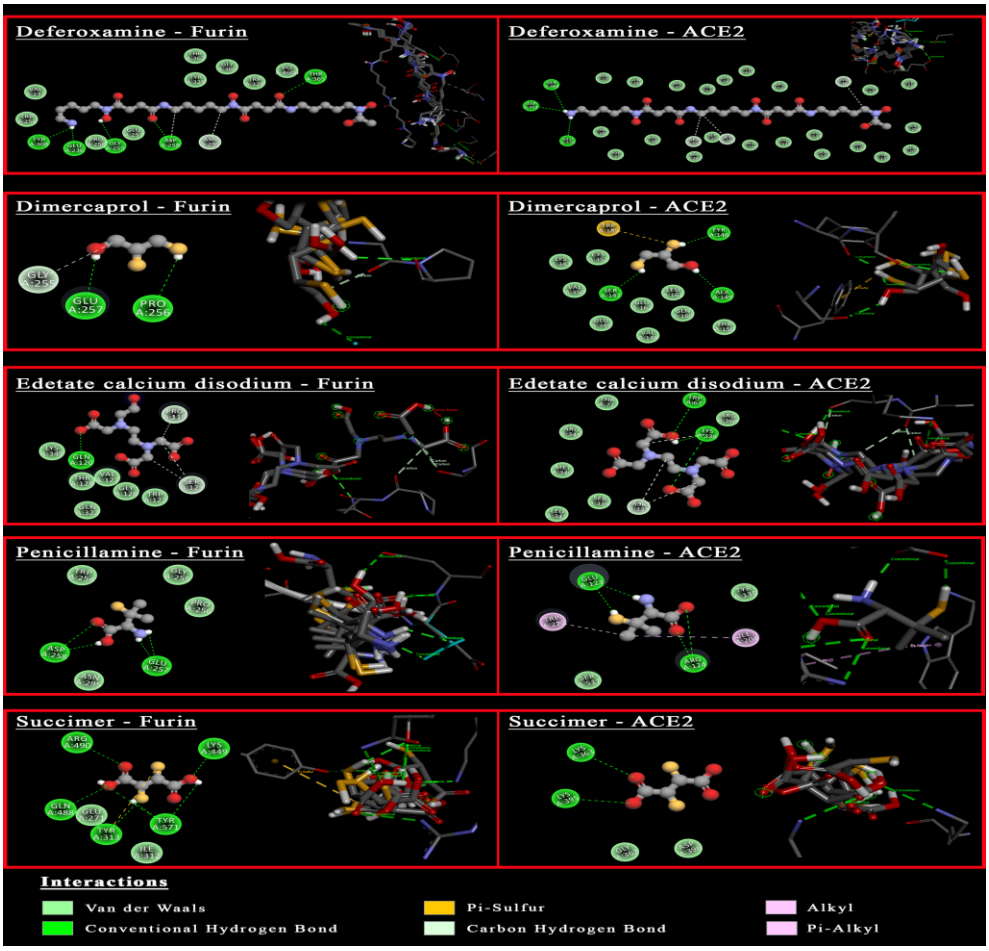

Figure 2. Docking analysis of the molecular interactions and the binding mode conformation of the calcium chelating compounds with ACE2 and Furin target proteins.

Molecular docking result showed that Deforexamine and Edetate calcium disodium display highest number of interaction with furin and ACE2 both. All though based on binding energy of Penicillamine and Dimercaprol displayed stable interaction with ACE2 and furin. Out of the five drugs, Penicillamine and Dimercaprol were potential drug candidates represented highest dock scores and strong interactions with catalytic active residues of Furin and ACE2 both (Fig 2). Although the toxicokinetics and half life are very different when compare Penicillamine with Dimercaprol. Penicillamine has less toxicological effect and has a much longer half-life up to 4-6 days, in case of long-term therapy.

Conclusion

The disease severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is spreading rapidly throughout the world. The gene of the SARS-CoV-2 spike protein adopt 12 stable nucleotides and the corresponding peptidal region of S2 region of the spike protein may reveal a screened set of epitopes that can guide to design an experimental effort towards the development of vaccines against SARS-CoV-2. Molecular docking resulted that Penicillamine and Dimercaprol compounds showed good inhibitory activity against Furin and ACE2. We therefore speculate that based on pharmacokinetics and toxicokinetics index, Penicillamine can be more effective to inhibit two receptor proteins of the host where the spike glyco-protein of SARS-CoV-2 binds.

References and notes

1. Lu, R., Zhao, X., Li, J., Niu, P., Yang, B., Wu, H., Wang, W., Song, H., Huang, B., Zhu, N., et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. Lancet. pii: S0140-6736(**20**)30251-8 (2020).

2. Menachery, V.D., Graham, R.L., and Baric, R.S.. Jumping species-a mechanism for coronavirus persistence and survival. Curr Opin Virol **23**, 1-7 (2017).
3. International Committee on Taxonomy of Viruses, ICTV. Virus Taxonomy: 2018b Release. URL: https://talk.ictvonline.org/taxonomy/. Revised on: February 9th, (2020).
4. Li F. Structure, Function, and Evolution of Coronavirus Spike Proteins. Annu Rev Virol **3**:237-261 (2016).
5. Li WH, Moore MJ, Vasilieva N, Sui JH, 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 **426**:450-454 2003.
6. Hajdin, K., D'Alessandro, V., Niggli, F. K., Schafer, B. W. & Bernasconi,M. Furin targeted drug delivery for treatment of rhabdomyosarcoma in a mouse model. PLoS. One. 5, e10445 (2010).
7. Thomas, G. Furin at the cutting edge: from protein traffic to embryogenesis and disease. Nat. Rev. Mol. Cell Biol. **3**, 753–766 (2002).
8. Xu X, Chen P, Wang J, Feng J, Zhou H, Li X, Zhong W, Hao P Evolution of the novel coronavirus from the ongoing Wuhan outbreak and modeling of its Spike protein for risk of human transmission. Sci China Life Sci. https://doi.org/10.1007/s11427-020-1637-5 (2020).
9. Lai, A.L., Millet, J.K., Daniel, S., Freed, J.H., and Whittaker, G.R. The SARS-CoV Fusion Peptide Forms an Extended Bipartite Fusion Platform that Perturbs Membrane Order in a Calcium-Dependent Manner. Journal of molecular biology. **429**. 3875-3892 (2017).
10. Basak A, Khatib AM, Mohottalage D, Basak S, Kolajova M, et al. A Novel Enediynyl Peptide Inhibitor of Furin That Blocks Processing of proPDGF-A, B and proVEGF-C. PLOS ONE 4(11): e7700. https://doi.org/10.1371/journal.pone.0007700 (2009).
11. Wanyiri JW, O'Connor R, Allison G, et al. Proteolytic processing of the Cryptosporidium glycoprotein gp40/15 by human furin and by a parasite-derived furin-like protease activity. *Infect Immun*.**75**(1):184–192. doi:10.1128/IAI.00944-06 (2007).
12. Belouzard, S., Millet, J.K., Licitra, B.N., and Whittaker, G.R. Mechanisms of coronavirus cell entry mediated by the viral spike protein. Viruses **4**, 1011-1033 (2012).
13. DeLano WL. Pymol: an open-source molecular graphics tool. CCP4 Newsl Protein Crystallogr;**40**:82–92 (2002).
14. Morris GM, Huey R, Lindstrom W, Sanner MF, Belew RK, Goodsell DS, et al. AutoDock4 and AutoDockTools4: automated docking with selective receptor flexibility. J Comput Chem. ;**30**:2785–91 (2009).
15. Andersen, K.G., Rambaut, A., Lipkin, W.I. *et al.* The proximal origin of SARS-CoV-2. *Nat Med* (2020).https://doi.org/10.1038/s41591-020-0820-9 (2020).

Acknowledgements: This work was supported by Oriental Institute of Science and Technology in the frame of our Research Programme.

Figure legend

Figure 1. Sequence comparison and structural deviation of the spike glyco-protein of SARS-CoV-2 binds. Newly inserted Nucleotides highlighted as yellow and green colour, the corresponding amino acids are in Red colour. The structures of spike glyco-proteins are modelled and structural deviation (RMSD) are presented.

Figure 2. Docking analysis of the molecular interactions and the binding mode conformation of the calcium chelating compounds with ACE2 and Furin target proteins.

Author information

Affiliations
Post Graduate Department of Biotechnology, Molecular informatics Laboratory, Oriental Institute of Science and Technology, Vidyasagar University, Midnapore, West Bengal, India.
Anindya Sundar Panja and Aniket Sarkar

Author contributions: A.S.P designing experiments and analyzing data; Method development, material preparation and experiments performed by A.S and A.S.P. interpreted the results.
Conflict of interest: The authors declare that they have no conflict of interest.