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STRUCTURE BASED DRUG DESIGN FOR SWINE FLU: SURFACE ANTIGEN NEURAMINIDASE AS A TARGET ON *IN SILICO* PLATFORM

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ABSTRACT

Swine flu (also known as seasonal flu or hog flu or swine influenza), is caused by Influenza A virus of various subtypes i.e. H1N1, H2N2, H3N2, H1N2 in both human and swine. One of the surface protein of Influenza virus, neuraminidase play an important role in the infection of progeny virions inside the respiratory tract of host organism. Present study is a *de novo* research work to predict possible neuraminidase inhibitor(s) with the help of bioinformatics software and online available tools. In this work, three dimensional tertiary structure of the neuraminidase protein has been predicted due to the absence of its structure in Protein Data Bank. The active site of the predicted model has been predicted using online computational tool and then allowed for docking studies. One of the best docking results has been obtained with Indacaterol, having minimum free energy of -8.2 kcal/mol.

Keywords: Neuraminidase, Protein Data Bank, Hog Flu, Swine flu, molecular docking, Indacaterol

INTRODUCTION

Influenza is one of the most common infectious viral disease which is caused by influenza virus of various types, mainly Influenza A, Influenza B and Influenza C in swine, avians and humans. Swine influenza (SI) is mainly caused by Influenza A of subtypes H1N1, H1N2, H2N2, H3N2 in both swine and humans (2). These subtypes

represent surface proteins Hemagglutinin (H) and Neuraminidase (N), possessing various roles in causing viral infections. Like other influenza, initially, SI is also characterized by myalgia, fever, headache, severe malaise, nonproductive cough, sore throat, and inflammation in lungs. During critical illness, it may cause death of the patient (4).

Influenza virus contain two glycoproteins i.e. hemagglutinin (HA) and neuraminidase (NA) on its surface which play an important role in the initiation of viral infection (1, 3). Hemagglutinin plays a significant role in infection with the target cell surface as it is present on the surface of the virion. Neuraminidase cleaves sialic acid (neuraminic acid) from the ends of the polysaccharide chains of receptors, to prevent the self aggregation of the progeny virions. Hence, neuraminidase enzymatic activity promotes the dispersement of influenza virus in the mucosal secretion within infected person (5, 6).

In the present study, neuraminidase surface protein has been targeted on in silico platform to synthesize a *de novo* drug. The study has followed the standard protocol and online databases for creating a computational model of the drug targeted against surface protein neuraminidase with significant role in swine influenza pathology.

METHODOLOGY

Sequence Alignment

The sequence of neuraminidase (query protein) has been obtained from the online resource National Centre for Biotechnology Information, NCBI (http://www.ncbi.nlm.nih.gov) in FASTA format. The sequence alignment was performed by using BLASTp, sequence alignment tool (http://blast.ncbi.nlm.nih.gov/Blast.cgi). The Chain A, Crystal structure of H2N2 neuraminidase of Influenza A virus (PBD ID: 3TIA) showed maximum identity and has been selected as a suitable template protein to create a three dimensional model of the target protein sequence.

Structure Prediction

Computational secondary structure of neuraminidase has been predicted followed by alignment with available sequences of protein by using an online server 3DPSSM and PHYRE2. Aligned results provided nine protein templates which were showing maximum identity with our query protein structure. With the help of Protein Data Bank, a computational database, PDB file format of the selected eight templates having resolution <3.0, R-Value <5.0 and X-Ray crystallographic structure, has been obtained. EASY MODELLER (version 4.0), has been

used to predict the 3-dimensional model of query protein, by submitting PDB file formats of the selected template protein.

Validation of Predicted Model (Loop Modeling)

Structural validation of the query protein model has been done with the help of the online server, Structural Analysis and Verification Server, SAVES (Prochek). The tool is capable in Ramachandran Plot analysis of the submitted protein model. The final structure obtained, has been viewed with Swiss PDB Viewer (SPDB 4.10) software.

Prediction of Active Site

Dog Site Scorer, an online server has been used to predict the active site of the submitted protein model and then, that active site can be used as a target for the *in silico* designed potential drug molecule. The active site of the query protein, having 469 amino acids has been predicted for the purpose.

Drug Library

Online drug databases like Drug Bank, has been used to study about and download the chemical structure of the drugs. In this work, Drug Bank database has been used to download the structure of various molecules available for Swine influenza. These downloaded molecules have been selected for the molecular docking studies.

Molecular Docking

Auto Dock is the online software, used for molecular docking. With the help of this software, docking studies with the downloaded chemical drug structure was performed against the targeted active site of the query protein and the compound that showed minimum free energy has been selected for further studies.

Results and Discussion

The sequence alignment of neuraminidase with BLASTp has resulted in 96% identity with the Chain A, Crystal structure of the neuraminidase of H2N2 subtype of Influenza A virus (Fig. 1). The computational model of neuraminidase has been predicted with the help of EASY MODELLER. The minimum dope score obtained for the template proteins corresponding to 32288-09768 (Fig. 2), with studies using 3DPPSM and PHYRE2. The Ramachandran Plot analysis of the query protein model has given the final protein structure of neuraminidase (Fig. 3 and Fig. 4) which has been viewed with Swiss PDB Viewer (Fig. 5). The results of active site analysis with the server, DogSite Scorer, have showed the active site of the query protein (Fig. 6) which has been used for docking studies. The molecule Indacaterol, corresponded to the minimum free energy value of -8.2 kcal/mol (Fig. 7).

CONCLUSION

Present study with *in silico* approach for *de novo* drug designing against swine influenza has been encouraging using various bioinformatics software and online servers we have predicted a three dimensional protein model of neuraminidase protein, its active site and a molecule that has showed minimum free energy with the active site binding. For further study, the molecule Indacaterol and its derivatives can be useful molecule for drug development.

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REFERENCES

[1] Manoj M Narayanan, Chandrasekhar B Nair, Shilpa K Sanjeeva, PV Subba Rao, Phani K Pullela, Colin J Barrow (2013). Design of multiligand inhibitors for the swine flu H1N1 neuraminidase binding site. Advances and Applications in Bioinformatics and Chemistry. 6:47-53.

[2] Ravi Patel, Ishan Panchal, Dilip Chavda, Priya Modiya, Hiren Marvaniya, Kaumil Modi and Dhrubo Jyoti Sen (2010). H1N1/09 Influenza: 20th Century Flu Pandemics. Journal of Chemical and Pharmaceutical Research. 2(3):605-619.

[3] Larisa V. Gubreva (2004). Molecular Mechanism of Influenza Virus Resistance to Neuraminidase Inhibitors. Virus Research. 103:199-203.

[4] Andrea G. Winquist, Keiji Fukuda, Carolyn B. Bridges, Nancy J. Cox (1999). Neuraminidase Inhibitors for Treatment of Influenza A and B Inhibitors. U.S. Department of Health and Human Services. Centre for Disease Control and Prevention.

[5] Elspeth Garman and Graeme Laver (2005). Chapter 17: The Structure, Function and Inhibition of Influenza Virus Neuraminidase. Viral Membrane Proteins: Structure, Functions and Drug Design. 247-267.

[6] Mark von Itzstein (2007). The war against Influenza: Discovery and Development of Sialidase Inhibitors. Nature Publishing Group. 6:967-974.

Fig. 1 Result of BLASTp of Neuraminidase Protein

Filename	MOL pdf	DOPE score	GA341 score
query.B99990001.pdb	22869-35352	32288-09768	0.9768
query.B99990002.pdb	22107-31836	32962-35156	0.08939
query.B99990003.pdb	22506-86914	33878-95312	0.99976
query.B99990004.pdb	22537-02539	33081-80078	0.06953
query.B99990005.pdb	23476-14648	32210-22266	0.29889
query.B99990006.pdb	23476-95117	30143-03125	0.07157
query.B99990007.pdb	22682-10938	32724-66211	0.11394
query.B99990008.pdb	22588-34375	32302-80469	0.11702
query.B99990009.pdb	23233-68164	32247-36328	0.11709

Fig. 2: Dope Score of the template protein molecules obtained after running Easy Modeller

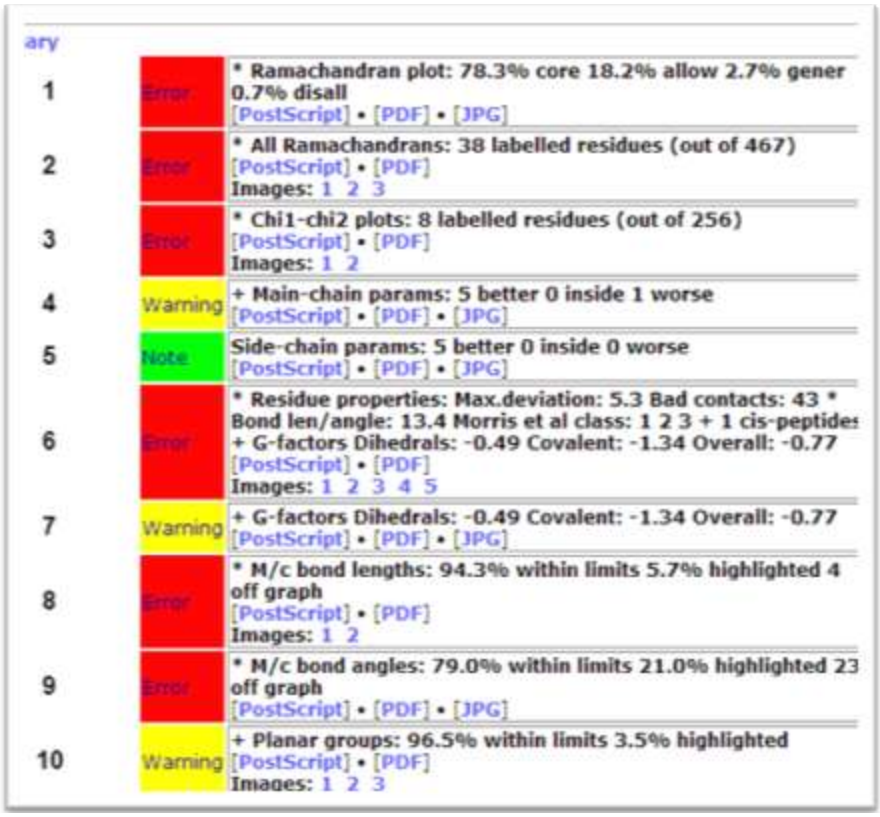

Fig.3: Results of Validation of Query Protein Model in SAVES (Prochek)

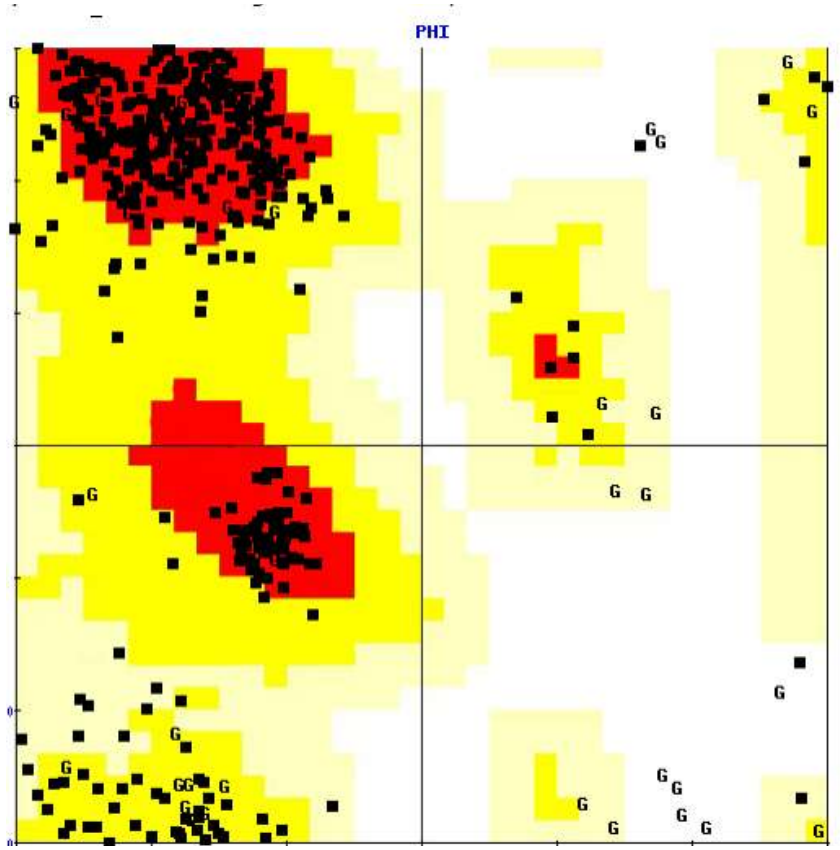

Fig. 4: Ramachandran Plot Analysis of Query Protein by SAVES (Prochek)

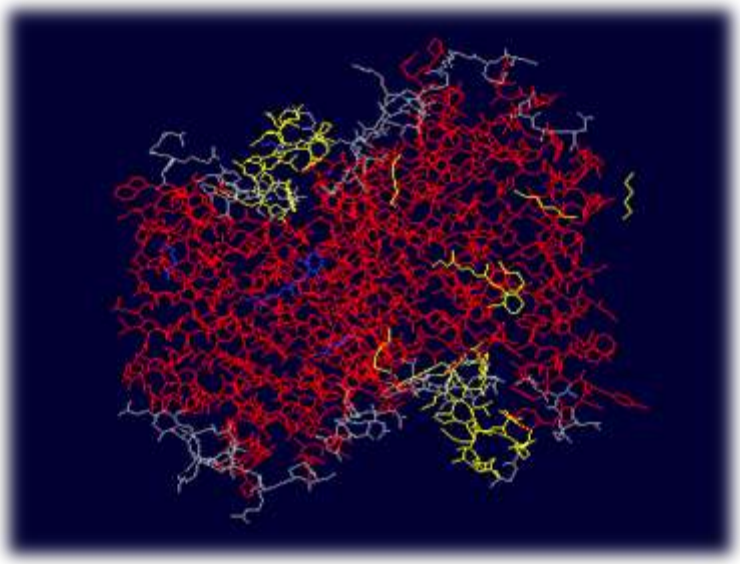

Fig. 5: Three dimensional structure of Neuraminidase protein viewed by Swiss PDB Viewer

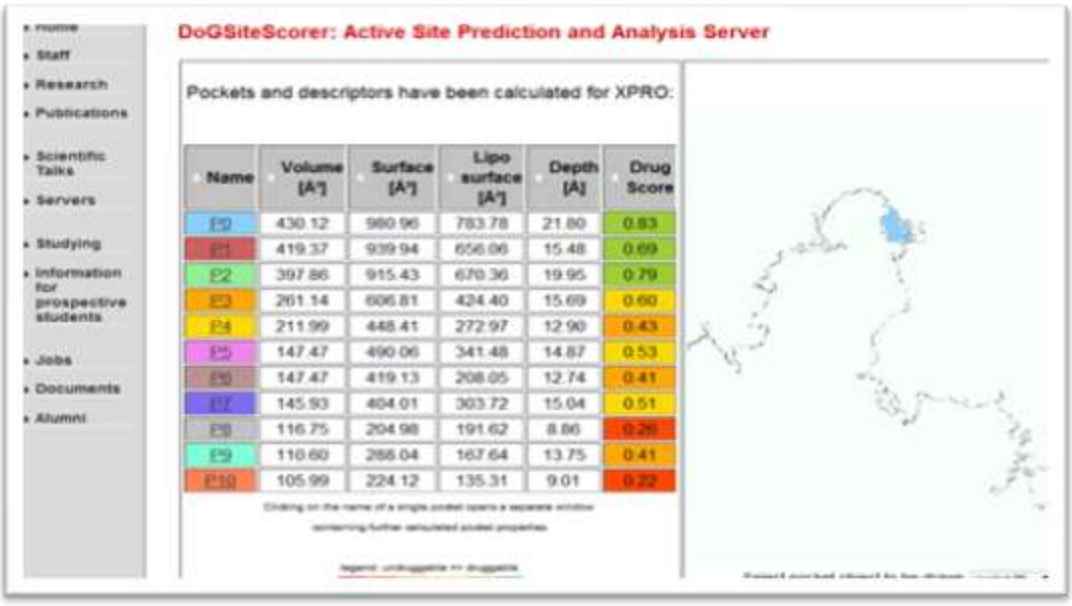

Fig. 6: Result of Dog Site Scorer, Active Site of protein showed in sky blue color

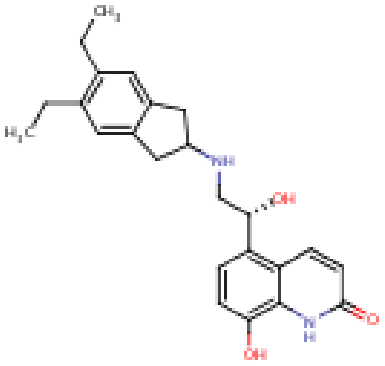

Fig. 7: Chemical Structure of Indacaterol