Journal of Materials Science & Surface Engineering, 6(6): 884-887
ISSN (Online): 2348-8956; 10.jmsse/2348-8956/6-6.2

In Silico Molecular Docking of Inhibitors Against Human Cytomegalovirus IE1: Protein responsible for Brain Tumour

Meenu Agarwal · Ayushi Verma

Department of Biotechnology, Noida Institute of Engineering and Technology, Greater Noida, U.P., India.

ABSTRACT

The IE1 protein modifies the brain tumour cells in such a way that these tumorous cells start growing at fast pace, these facts could establish a conformed performance of these cells for CMV in the "malignant process of this tumour". CMV Cytomegalovirus(CMV) is**a** typical human virus contains a typical presence of a protein that plays a vital role in turning the human brain tumours more aggressive and spreading with fast pace.In the present study six potential inhibitors (HDAC, ACE, Bromelain, creatinine and benzo resorcinol, GSK) were selected against IE1protein for docking analysis. Interaction between IE1 protein and inhibitors was done using Hex 6.1 version, PatchDock and FireDock. Docking analysis revealed that GSK inhibitor has shown minimum binding energy (-775.50) and maximum score (1060). Thus, proving to be a strong inhibitor against IE1 protein.Further GSK can be considered as a potential lead compound for drug designing against tumour cells.

ARTICLE HISTORY
Received 19-03-2019
Revised 03-04-2019
Accepted 04-04-2019
Published 01-12-2019

KEYWORDS
Docking
PDB
NCBI
Inhibitors

Introduction

A brain tumour is a mass of abnormal tissue growing in any part of the brain. For some unknown reason, some brain cells multiply in an uncontrolled manner and form these tumours. These tumours can arise from any part of the brain, spinal cord or the nerves. Broadly these tumours can be divided into Benign and malignant tumours. Benign tumours grow slowly and never spread to other parts. But as they slowly increase in size they can cause pressure on the normal brain and interfere with mental and bodily functions. Some of the benign tumours known are: meningioma's, pituitary adenoma, craniopharyngioma, epidermoid cysts, neurocytoma, haemangioma, pilocytic astrocytoma, etc [1, 2].

Malignant tumours are aggressive tumours that grow fast and infiltrate the surrounding brain and sometimes spread to the other parts of the brain or spine. There are various types of malignant brain tumours like High Grade Astrocytoma/Glioma, ependymoma, PNET, medulloblastoma, lymphoma, Germ cell tumours with aggressive and timely treatment some of these can be cured.

"Cytomegalovirus", a typical human virus contains a typical presence of a protein that plays a vital role in turning the human brain tumours more aggressive and spreading with fast pace. It was found out that Human cytomegalovirus(HCMV) immediate early (IE)proteins that are endogenously expressed in GBM cells are strong viral trans activators with oncogenic properties. It modifies the brain tumour cells in such a way that these tumorous cells start growing at fast pace, these facts could establish a conformed performance of these cells for CMV in the "malignant process of this tumour".

Tumour cells receive a 'two-punch' blow by the "IE1" protein. It is found that this 'CMVIE1' protein in almost every malignant brain tumour [3]. This spotting led to the fact that "it can make tumour cells more aggressive by affecting two major pathways".

The process has two stages. The first stage is for inhibiting two key tumour suppressing proteins within the brain tumour cells. During the second stage, there is a promotion of growth with pathway signals in the tumour itself. When both these cellular pathways are modulated the tumour cells get more agitated by the viral protein. CMV, a very common virus, is one of the main causes for persisting infection in almost 50% to 60% of the Americans. This disease displays very few symptoms and people at large are not much aware of these symptoms of infection [4].CMV could be a great risk factor even for the healthy individuals who have compromised immune system. Such a situation may be common with the people suffering through HIV/AIDs along with the organ transplant aspirants, and the pregnant women with their fetus.

A substance used in the diagnosis, treatment, or prevention of a disease or as a component of a medication is called as drug. Rational drug design (RDD) is a process used in the biopharmaceutical industry to discover and develop new drug compounds. RDD uses a variety of computational methods to identify novel compounds, design compounds for selectivity, efficacy and safety, and develop compounds into clinical trial candidates [5]. These methods fall into several natural categories – structure-based drug design, ligand-based drug design and de novo design– depending on how much information is available about drug targets and potential drug compounds. The ideal situation in RDD is to have knowledge of the target protein structure along with bound ligands. The more available knowledge, the better the chances of designing and optimizing ligands to modulate therapeutic targets. But there can still be surprises, including alternative modes of ligand binding and conformational changes in the target protein structure. Rational approaches can advance drug discovery even when the target protein structure is not known. The structure of at least one active ligand can be used to identify new ligands [6].

In the modern drug discovery process, RDD means efficiently using information from all possible sources to

Corresponding Author: Meenu Agarwal, Tel: +91 7065556698
Email: meenuagrawal06@gmail.com

Contents lists available at http://www.jmsse.in/ & http://www.jmsse.org/

prioritize the next steps, elucidate trends, rank compounds, and avoid unproductive experiments. Rational approaches encompass target protein structure-based design of ligands, design of novel ligands based on known ligands, and consideration of the drug-like properties of lead molecules in addition to their potency and selectivity. In vitro experiments and in silico modelling, screening, and prediction can all generate data in a high-throughput manner. Storing, tracking, and using all of the data to make decisions are challenges for building informatics platforms.
Structure-based drug design is one of several methods in the rational drug design toolbox. Drug targets are typically key molecules involved in a specific metabolic or cell signalling pathway that is known, or believed, to be related to a particular disease state. Drug targets are most often proteins and enzymes in these pathways. Drug compounds are designed to inhibit, restore or otherwise modify the structure and behaviour of disease-related proteins and enzymes. SBDD uses the known 3D geometrical shape or structure of proteins to assist in the development of new drug compounds. The 3D structure of protein targets is most often derived from x-ray crystallography or nuclear magnetic resonance (NMR) techniques. X-ray and NMR methods can resolve the structure of proteins to a resolution of a few angstroms (about 500,000 times smaller than the diameter of a human hair). At this level of resolution, researchers can precisely examine the interactions between atoms in protein targets and atoms in potential drug compounds that bind to the proteins.
This ability to work at high resolution with both proteins and drug compounds makes SBDD one of the most powerful methods in drug design.
Bioinformatics plays an important role in the design of new drug compounds. The input of Biocomputing in drug discovery is two folds: firstly, the computer may help to optimize the pharmacological profile of existing drugs by guiding the synthesis of new and "better" compounds. Secondly, as more and more structural information on possible protein targets and their biochemical role in the cell becomes available, completely new therapeutic concepts can be developed. The computer helps in both steps: to find out about possible biological functions of a protein by comparing its amino acid sequence to databases of proteins with known function, and to understand the molecular workings of a given protein structure. Understanding the biological or biochemical mechanism of a disease then often suggests the types of molecules needed for new drugs. In the present study, we are trying to find out best inhibitor molecule by their energy optimization for IE1protein. The main objectives of this study are to retrieve the protein sequence of IE1protein, to retrieve the structure of IE1 protein, to perform docking of this protein with its inhibitors by using Hex 6.1 version docking program, PatchDock & FireDock. Further evaluation of the docking results will find the inhibitor having maximum interaction with the IE1 protein.

Experimental

Materials and Methods

Sequence retrieval

The 3d structure of protein was obtained from protein data bank (PDB).
PDB: The PDB provides a variety of tools and resources for studying the structures of biological macromolecules and their relationships to sequence, function, and disease. This site offers tools for browsing, searching, and reporting that utilize the data resulting from ongoing efforts to create a more consistent and comprehensive archive.

Structure of IE1 protein

The following steps were involved in the retrieval of structure of IE1 protein:

1. CID no.1T18 was selected among the list of protein displayed in the search result in Protein Data Bank(PDB).
2. From the option 'Download PDB file', the PDB file was saved in Microsoft word. This PDB file was further used in docking procedures.

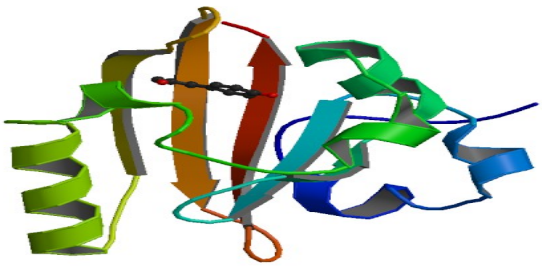


Figure 1: 3-D Structure of IE1 Protein

Retrieval of inhibitors

The inhibitors for IE1 protein enzyme were obtained from PubChem database of NCBI, DRUG BANK etc. These databases give all the information about the inhibitors, such as their structure, IUPAC name, molecular weight (Table 1).

NCBI PubChem database:

It is a database/collection of all the chemical compounds, drugs, ligands etc which are directly or indirectly associated with metabolism or physiology of living organisms.

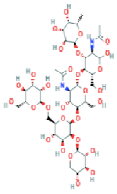
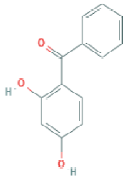
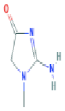
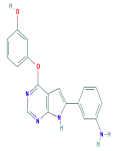
Docking of ligand to receptor

For Docking of ligand to receptor, programs such as AutoDock, Hex, GRAMM, GLIDE, FLEXX, GOLD, HINT, PatchDock, FireDock etc are used.

Hex tool

Hex is the only docking and superposition program to use spherical polar Fourier (SPF) correlations to accelerate the calculations, and its still one of the few docking programs which has built-in graphics to view the results. Also, it is the first protein docking program to be able to use modern graphics processor units (GPUs) to accelerate the calculations.*Hex* is an interactive molecular graphics program for calculating and displaying feasible docking modes of pairs of protein and DNA molecules. *Hex* can also calculate protein-ligand docking, assuming the ligand is rigid, and it can superpose pairs of molecules using only knowledge of their 3D shapes. In *Hex*'s docking calculations, each molecule is modelled using 3D expansions of real orthogonal spherical polar basis functions to encode both surface shape and electrostatic charge and potential distributions.
Essentially, this allows each property to be represented by a vector of coefficients (which are the components of the

Table 1: PubChem CID, 2-D Structures of Inhibitors

S.NO	INHIBITORS	PUBCHEM CID	2-D STRUCTURE	IUPAC	MOLECULAR WEIGHT	MOLECULAR FORMULA
1	HDAC	5311		N-Hydroxy-N'-phenyloctanediamide	264.33 g/mol	$C_{14}H_{20}N_2O_3$
2	ACE	11074431		6-Methyl-2,2-dioxooxathiazin-4-olate	201.24 g/mol	$C_4H_4KNO_4S$
3	Bromelain	44263865		*N*-[2-[5-acetamido-6-hydroxy-2-(hydroxymethyl)-4-(3,4,5-trihydroxy-6-methyloxan-2-yl)oxyoxan-3-yl]	26.6 kDa	$C_{39}H_{66}N_2O_{29}$
4	Benzoresorcinol	8572		2,4-Dihydroxybenzophenone	214.21g/mol	$C_{13}H_{10}O_3$
5	Creatinine	588		2-amino-3-methyl-4*H*-imidazol-5-one	13.3 k Da	$C_4H_7N_3O$
6	GSK	9549289		3-[[6-(3-Aminophenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-yl]oxy]phenol	318g/mol	$C_{18}H_{14}N_4O_2$

basis functions). *Hex* represents the surface shapes of proteins using a two-term surface skin plus van der Waals steric density model, whereas the electrostatic model is derived from classical electrostatic theory.

Hex version 6.1

1D and 3D calculations may optionally be performed on one or more GPUs, which can give a considerable speed-up compared to using conventional CPUs. In this case (also counter-intuitively), 1D correlations are much faster than 3D correlations on the GPU. Additionally, the CPU-based calculations have also been re-written to use multi-threading in order to support parallelisation on both Windows and Linux-based multi-core systems. Thus, significant performance improvements can be expected from version 6.1.

PyMOL

PyMOL is an open-source model visualization tools available for use in structural biology.It is well suited to produce high quality 3D images of small molecules and biological macromolecules such as proteins. The 'Py' portion of the software's name refers to the fact that it extends, and is extensible by, the Python programming language.

The following steps were involved in this procedure.

1. The file with extension .pdb, (which was formed in hex procedure), was opened.
2. This file was copied and pasted at the bottom of the protein .pdb file and saved this as PDB file.
3. PYMOL was opened, 'File' menu was clicked and PDB file was opened.
4. 'Action', 'preset', 'ligand sites', 'solid (better)', menu was clicked, this showed surface structure of protein with bound inhibitor.
5. This file was saved with the extension '.png'.

Results and Discussion

IE1protein, which is responsible for brain tumour and it's inhibitors can be widely used as a drug target. Many inhibitors are also known to have this protein as their target.1T18 is the CID of the IE1 protein. For finding the inhibitor of IE1 with best interaction with the protein, the IE1protein was docked with a list of the inhibitors known to interact with it. After retrieving the sequence and then structure of the protein, it was docked with its inhibitors

Table 2: Results displayed by HEX docking program displaying docked energy of the IE1with its inhibitor

S.No	Inhibitor	PubChem CID	Minimum docked energy
1.	HDAC	5311	- 582.01
2.	ACE	11074431	-487.30
3.	Bromelain	44263865	-691.39
4.	Benzoresorcinol	8572	-484.81
5.	Creatinine	588	-570.93
6.	GSK	9549289	-775.50

Table 3: Results displayed by PatchDock &FireDock displaying score and Global energy of the IE1with its inhibitor

S.No	Inhibitor Name	PubChem CID	PatchDock (SCORE)	FireDock (Global Energy)
1	HDAC	5311	762	-6.99
2	ACE	11074431	1060	-86.50
3	Bromelain	44263865	822	-52.25
4	Benzoresorcinol	8572	874	-43.50
5	Creatinine	588	1060	-72.52
6	GSK	9549289	1060	-183.02

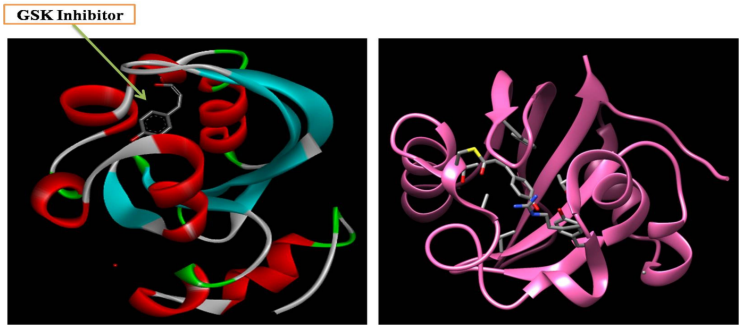


Figure 2: Insilico binding of Inhibitor GSK with IE1 Protein with binding energy -775.50 using (A)Discovery Studio 4.2 and (B) Chimera Software

and the inhibitor with best docking results provides best interaction with the IE1 protein.Docking is done with the help of Hex 6.1,PatchDock & FireDock .Further the docking result analysis have proved GSK to be the best inhibitor among other inhibitors to inhibit IE1 protein because it has low docked energy which signifies better interaction of ligand with the protein (Table 2 & Table 3).

Conclusions

Pharmaceutical companies prefer to use best docking software, so as to design new drugs with excellent efficacy and with low price costs. Rational drug designing strategies helps to find out the best inhibitor for any disease and also reduces the cost of the drugs for the disease. Therefore, this area provides vast opportunities in research and designing of drugs. IE1protein was modelled and known inhibitors of the protein were identified. In the present work, we found that GSK is the best selective inhibitor (in terms of stability of the complex) of IE1 among different types of inhibitors because it shows maximum free energy (in -ve). Interaction of the inhibitors with the protein was examined through Insilco Docking Approach.

The rationale of the present work is amply justified by the presented results. From docking studies, a suitable inhibitor GSK is hunted which can further prove to be the best inhibitor to inhibit the IE1protein . The current work on inhibitor optimization could be a platform for the further studies in drug designing for most cost effective, time consuming effective approach for finding the inhibitor against this brain tumour.

References

1. Yagub Allen, T.M. Tat-specific cytotoxic T lymphocytes selected for SIV escape variants during resolution of primary viraemia. Nature, 2000, 407, 386-390.
2. Bedell M.A., Jones K.H., and Laimins L.A. The E6-E7 region of human papillomavirus type 18 is sufficient for transformation of NIH 3T3 and rat-1 cells. Journal of Virology, 1987, 61, 3635-3640.
3. Brown DR, Fan L, Jones J, and Bryan. Colocalization of human papillomavirus type 11 E1^E4 and L1 proteins in human foreskin implants grown in athymic mice. Journal of Virology, 1994, 201, 46-54.
4. Butel, J. Viral carcinogenesis: revelation of molecular mechanisms and etiology of human disease. Carcinogenesis, 21, 405-426.
5. Nagpal J.K., Sahni S., Das B.R. p53 codon 72 polymorphism and susceptibility to development of human virus-associated cervical cancer in Indian women. Europian Journal Clinical Investigation. 2002, 32, 943-948.
6. Khare S., Pater M.M., Tang S.C., and Pater A. Effect of glucocorticoid hormones on viral gene expression, growth and dysplastic differentiation in HPV 16-immortalized ectocervical cells. Experimental Cell Research, 1996, 232, 353-360.