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Research Article

In-silico Studies of Quinazolinone Analogues to Distinguish their Hypothetical Binding Mode using the X-ray crystal Structure Human carbon Anhydrase II (HCAII) Enzyme Complex with Sugar Sulfamate for Anticonvulsant Activity

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ABSTRACT

The quinazolinone moiety is a significant pharmacophore that depicts various types of pharmacological activities as shown in recent exhaustive ligatures. Quinazolinone exhibit potent central nervous system (CNS) activities like anti-anxiety, analgesic, anti-inflammatory and anticonvulsant. To develop these views and application profiles, attempt have been made to report a drug/ligand or receptor/protein interactions by identifying the suitable active site against X-ray crystal structure of Human Carbonic Anhydrase II (HCA II) enzyme for anticonvulsant activity using Vlife MDS version 4.6 Software because the protein-ligand interaction plays a significant role in structural based drug designing. The interaction was evaluated based on the score comparison between quinazolinone derivatives with sugar sulfamate. The quinazolinone ring forms hydrophobic and hydrogen bond contacts amino acid residues. The ligands 4t and 4s were shown to possess minimum dock score *i.e.* minimum binding energy in Kcal/mole *i.e.* these molecules has more affinity for the active site of the receptor. Molecules with low dock score and binding energy show more affinity towards the receptor. The data reported in this article may be helpful for the medicinal chemists who are working in this area.

Introduction

Epilepsy is a chronic brain disorder that dramatically affects people of all ages. It is characterized by spontaneous recurrent seizures that are related to a rapid change in ionic composition, including an increase in intracellular potassium concentration and pH shifts. People with epilepsy and their families can still be targets of stigma and prejudice today with consequent social discrimination. This is particularly evident in low and middle-income countries where 75% of the people affected do not receive the treatment they need because of economic as well as cultural circumstances. The high impact of the disease on global health has provoked immense efforts from the scientific community to shed light on the complex mechanisms underlying seizure generation and to develop therapeutic strategies to pharmacologically treat epilepsy. However, antiepileptic drugs (AED) currently available and employed in clinical practice can treat only some subtypes of epilepsy and, often, pharmacological treatment may not be resolute. For this reason, there is an urgent need to identify new molecular targets to expand the therapeutic options to treat and defeat this dramatic pathology. Carbonic anhydrases (CAs) are a group of ubiquitously expressed metalloenzymes that catalyze the reversible hydration/dehydration of CO_2/HCO_3. Thus, they are involved in those physiological and pathological processes in which cellular pH buffering plays a relevant role. It has been reported that CAs II, VII and XIV

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are implicated in epilepsy. In this context, selective CAIs towards the mentioned isoforms (CAs II, VII and XIV) have been proposed and exploited as anticonvulsant agents in treating epilepsy. The literature search in this field has turned highlighted on the role of carbonic anhydrases (CA) in epilepsy and it has emerged as an attractive target for designing new anticonvulsant drugs. The inhibition of CAs has pharmacologic applications for several diseases. In addition to the well-known employment of CA inhibitors (CAIs) as diuretics and antiglaucoma drugs, it has recently been demonstrated that CAIs could be considered as valid therapeutic agents against obesity, cancer, kidney dysfunction, migraine, Alzheimer's disease and epilepsy.[1-3]

The substituted quinazoline and quinazolinone derivatives shows an extensive range of biological activities such as anticancer, antimalarial, anticonvulsant, antiviral, antifungal, anti-protozoal, antimicrobial, anti-inflammatory, diuretic, muscle relaxant, antidepressant, anti-tubercular, and many other pharmacological activities.[4-11] Their easy synthetic accessibility and flexibility in structural modifications and functionalization of the same increases the importance of their appeal in medicinal chemistry. Molecular docking helps in the identification of the Ligands correct Binding geometry (Pose) within the binding site (Binding mode) and prediction of the binding affinity (Scoring Function) towards the receptor site.[12-20] In the present research work, we have docked quinazolinone analogs with X-ray crystal structure of human carbon anhydrase II (HCA II) enzyme complex with sugar sulfamate downloaded from PDB website. From the results obtained, I has been confirm that the ligands 4t and 4s were shown to possess minimum dock score *i.e.* minimum binding energy in Kcal/mole *i.e.* these molecules has more affinity for active site of the receptor (with X-ray crystal structure of human carbon anhydrase II (HCA II). Clearly indicates that molecules having low dock score and binding energy shows more affinity towards the receptor. The new examine the quinazolinone class of CAIs might represent novel therapeutic options for treating epilepsy.

Material and Methods

Molecular Docking

Molecular Docking studies and conformational analysis were performed by using the Molecular Design Suite (VLife MDS software package, version 4.6; from VLife Sciences, Pune, India).[23]

The compounds evaluated *in-silico* (docking) to distinguish their hypothetical binding mode using the X-ray crystal structure of human carbon anhydrase II (HCA II) enzyme complex with sugar sulfamate to possess the anticonvulsant activity behavior of designed ligands on structural basis. Docking studies were carried out and scoring functions (Figs. 1-4), their binding affinities and the orientation of designed compounds having (HCA II) antagonistic property.

Selection and Preparation of Protein

The 3 D structure of the receptor should be considered which is downloaded from PDB website. Firstly, all the water molecules were removed and hydrogen atoms were added, further by using the exploitation bioproducts tool, incomplete residues are completed in to the receptor. The co-crystallized matter cavity of a protein is identified. The energy of the complex obtained was decreased by using a force field. Chains of the receptor are separated and cavities were identified for docking.

Selection, Preparation and Optimization of Ligand

2D Structures of the selected compounds were drawn in to Vlife2Ddraw and converted to 3D structures further, it was saved in to mol2 format. All molecules were optimized by using AMBER method to reduce the energy of the molecules, taking the root mean square gradient (RMS) of 0.01 kcal/moleA° and the interaction limit to 10,000. Conformers for each structure were generated using Monte Carlo be applying AMBER force field method and the least energy conformer was selected for further study.

Molecular Docking

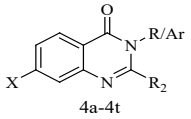
The 3D structures were energetically decreased up to the RMS gradient of zero.01 using MMFF. The cavities within the receptor were mapped to assign an appropriate active site, the fundamental feature used to map the cavities are the surface mapping of the receptor and identifying the geometric voids in addition as scaling the void for its hydrophobic characteristics. Hence, all the cavities present in receptor are identified and cavity no 1 was selected for docking based on their size and hydrophobic surface area having all atoms within a 5A° radius. Then, keeping all compounds for batch docking, the completion of batch docking would provide dock score. The interaction between ligand and receptor from a docked ligand-receptor complex is confirmed at the end.

Results and Discussion

Docking Studies

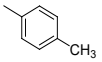
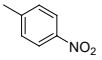
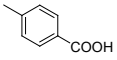
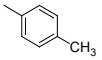
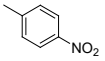
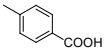
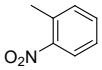
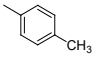
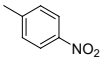
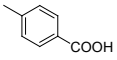
The docking results were evaluated in terms of docking score into the active site. Molecules with minimum dock score show more affinity for (HCA II) inhibition. The ligands 4t and 4s were shown to possess minimum dock score *i.e.,* minimum binding energy in Kcal/mole *i.e.*, this molecule has more affinity for active site of the residue of protein by the interaction of Hydrophobic bonds with protein residue, whereas the remaining compounds show less hydrophobic interaction, which could also be the probable reason for its lowest activity. The observations are given in Table 1.

Table 1: Docking score of the 2, 3- disubstituted-quinazolin-4(3*H*)-one using the X-ray crystal structure of HCA II) enzyme complex with sugar sulfamate for anticonvulsant activity



4a-4t

2, 3- disubstitutedquinazolin-4(3*H*)-ones

Compound No.	*Compound code*	*X*	*R2*	*R/Ar*	*Dock score (kcal/mole)*
1	4a	H	CH_3	H	-13.020367
2	4b	H	CH_3	NH2	-13.536103
3	4c	H	C_2H_5	H	-11.095004
4	4d	H	C_2H_5		-19.582934
5	4e	H	C_2H_5		-17.592799
6	4f	H	C_2H_5		-11.037822
7	4g	H	C_6H_5	H	-1.808366
8	4h	H	C_2H_5		7.656219
9	4i	H	C_2H_5		-17.906562
10	4j	H	C_2H_5	-NH2	-12.047577
11	4k	H	C_3H_7		-0.934317
12	4l	H	C_3H_7		0.637778
13	4m	H	C_3H_7		20.395301
14	4n	H	C_3H_7		-4.520501
15	4o	H	C_3H_7	-NH2	-24.717282
16	4p	Br	C_3H_7		-22.226013
17	4q	Br	C_3H_7		-5.043334
18	4r	Br	C_3H_7		-11.787338
19	4s	Br	C_3H_7		-28.928744
20	4t	Br	C_3H_7	$-NH_2$	-34.547319
Reference Molecule					-36.928555

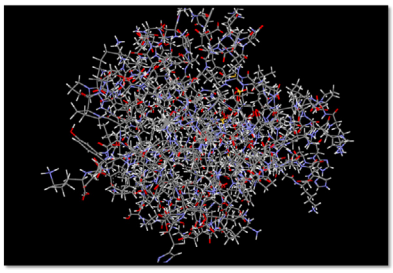


Fig. 1: X-ray crystal structure of human carbon anhydrase II (HCA II) enzyme

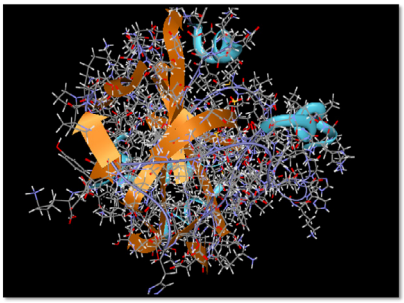


Fig. 2: X-ray crystal structure of human carbon anhydrase II (HCA II) enzyme (1eou) Assorted form

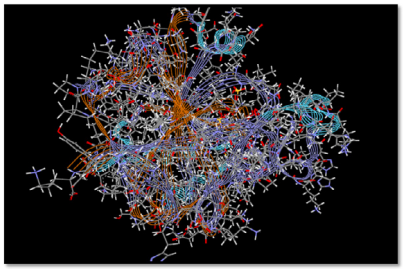


Fig. 3: X-ray crystal structure of human carbon anhydrase II (HCA II) enzyme (1eou) line strip form

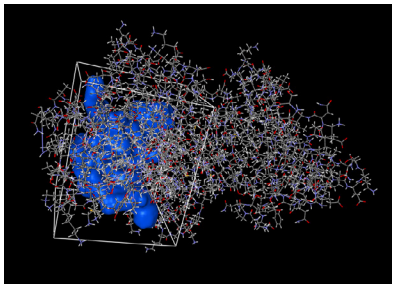


Fig. 4: The active site (Cavity) of 1eou receptor

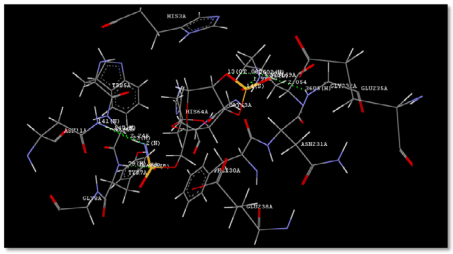


Fig. 5: The Reference molecule (sugar sulfamate) show the Hydrogen bonding

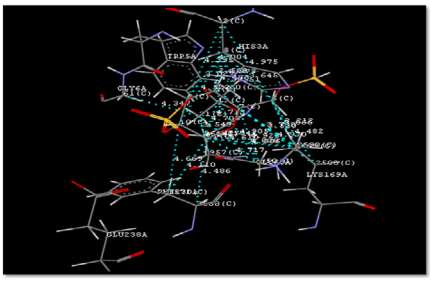


Fig. 6: The Reference molecule (sugar sulfamate) shows the hydrophobic bonding

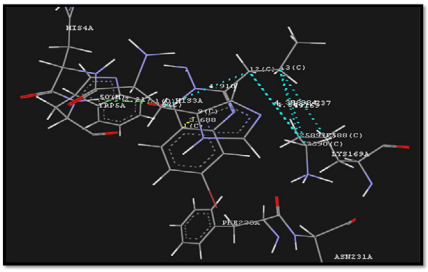


Fig. 7: The molecule (4t) shows the hydrogen, hydrophobic and pi Statching bonding

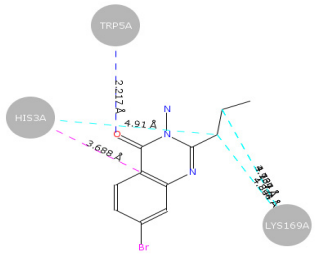


Fig. 8: 2D-view of the molecule 20 (4t) shows the hydrogen, hydrophobic and pi statching bonding

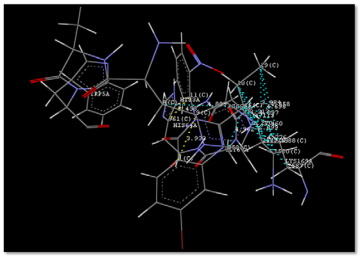


Fig. 9: The molecule (4s) shows the hydrophobic and Pi-Statching bonding

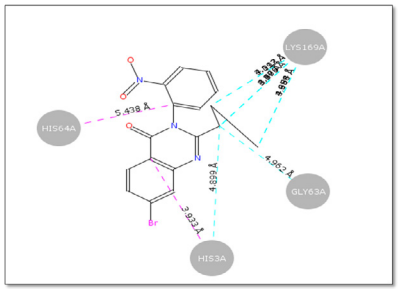


Fig. 10: 2D view of the molecule (4s) shows the hydrophobic bonding and Pi-Statching

Interactions of Ligands with Receptor

Reference molecule: sugar sulfamate

This is a reference molecule. It is HCA II antagonist. The dock score of this molecule is - 36.928555 kcal/mole (Figs. 5 and 6).

Molecule 20 (4t) The low dock score of this ligand is -34.547319 kcal/mole (Figs. 7 and 8).

Molecule (4s) The low dock score of this ligand is -28.928744 kcal/mole (Figs. 9 and 10)

Conclusion

The intermolecular interactions in between the ligand and the protein (receptor) were investigated using the MDS version 4.6, the 3-dimensional structure and the information about it was taken from PDB. 2, 3-disubstitutedquinazoline -4(3*H*) - one derivatives were docked into active site of receptor. A correlation was calculated by energy of binding premeditated by MDS.

The prediction of the potency or affinity of the ligand to the receptor was done by considering some parameters such as dock score, the energy of binding of molecules with the receptor, vander-waals interactions, H-bond interactions and charge interactions. More the negative value of the energy of binding the superior is affinity of the molecule to the receptor. Number of the vander-waals interaction shows that the ligand structure is having more number of bulky group due to which vander-waals interactions can be formed If the charge interactions are presents it helps verdict more suitable binding and so shows greater affinity to the receptor, contributing more potency.

MDS version 4.6, an automated docking programmed, successfully reproduced the binding mode of the X-ray crystal structure of HCA II enzyme for anticonvulsant activity. The ligands 4t and 4s were shown to possess minimum dock score *i.e.* minimum binding energy in Kcal/mole *i.e.* these molecules has more affinity for active site of receptor. Clearly, Molecules having low dock score and binding energy shows more affinity towards the receptor.

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