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Research Article

EVALUATION OF DRUG CANDIDATURE OF SOME QUINAZOLINE- 4-(3H)-ONES AS INHIBITOR OF HUMAN DIHYDROFOLATE REDUCTASE ENZYME: MOLECULAR DOCKING AND IN SILICO STUDIES

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ABSTRACT

Objectives: Human dihydrofolate reductase (hDHFR) is one of the best targets for the anticancer drug because it plays an important role in the synthesis of purines and pyrimidines. It also maintains intracellular biochemically active reduced folate pools. Quinazoline-containing compounds are more noticed because of their some resemblance with folic acid and also have provided attractive scaffolds for designing anticancer drugs. In this study, molecular docking and *In silico* studies were carried out in an attempt to evaluate the drug candidature of some quinazoline-4-(3H) -ones as inhibitors of human dihydrofolate reductase enzyme.

Methods: The study comprised of 27 compounds belonging to quinazoline-4-(3H)-one along with one standard drug methotrexate. Automated molecular docking of some quinazoline-4(3H)-ones with human DHFR was performed by the AutoDock 4.0 suite. Molecular descriptor properties were predicted by Molinspiration and OSIRIS Property explorer. Ligand based pharmacophore has been generated by PharmaGist tools.

Results: All the derivatives have qualified the Lipinski's Rule of Five and occupied the same cavity (as evidenced by the molecular docking results) in the protein molecule as is occupied by the natural ligand folic acid and the standard drug methotrexate. The binding energies of all the docked complex of compounds have significant negative values as compared to methotrexate.

Conclusion: The molecular docking study signified that the compounds can act as a putative inhibitor of hDHFR. The generated pharmacophore could further be used to design and develop new drugs. This study significantly supports a theoretical perception regarding the candidature of these compounds as inhibitors of human DHFR.

Keywords: Dihydrofolate reductase, Molecular docking, Drug likeliness, Drug score, Anticancer drug.

INTRODUCTION

In cancer chemotherapy, the folate metabolism has long been considered as an attractive target because of its obligatory role in the biosynthesis of nucleic acid precursor [1]. In folate metabolism, dihydrofolate reductase (5, 6, 7, 8 tetrahydrofolate: $NADP^+$ oxidoreductase, EC 1.5.1.3, DHFR) catalyzes the reduction of folate or 7, 8-dihydrofolate to tetrahydrofolate and intimately couples with thymidylate synthase. DHFR plays a fundamental role in the maintenance of intracellular biochemically active reduced folate pools [2]. It is also an important target for the treatment of a wide range of diseases. The abilities of quinazolines to inhibit DHFR activities were reported earlier [3-6].

Quinazoline-containing compounds have provided attractive scaffolds for designing anticancer drugs [7]. They are more noticed because of their diverse biological activity notably as kinase inhibitors [8] and some resemblance with folic acid. [9,10] The 4-anilinoquinazoline derivatives have led to the development and marketing of a new series of antitumor agents, such as gefitinib, erlotinib and lapatinib [11-13]. The quinazoline ring provides a satisfactory backbone for inhibition of mammalian DHFR, establishing contact with the key amino acid residues in the enzyme pocket. The 3-amino-2-aryl-4(3H)-quinazolinone was found to be highly potential against the multiple-antibiotic-resistant bacteria [14] and later antifungal and anti viral properties were also reported [15]. But literatures revealing anticancer property of these compounds are inadequate.

A suitable screening of the compounds, using theoretical and computational approaches prior to real-time experiments, to generate pharmacophore is considered to be the most appropriate strategy in the context of drug discovery research. The physico-chemical properties closely related to drug absorption are used in predicting bioavailability and also to interpret *in vitro* and *in vivo* findings. However, it is also found that the intrinsic biological and physiochemical parameters of the molecules depend on many of these properties. But, the complex structure of the whole drug molecule seems difficult to correlate with these parameters [16].

Additionally, modern drug design process helps to identify and develop new ligands with high binding affinity towards a target protein receptor. The molecular docking approaches help to reveal drug–receptor interaction to a greater detail. The study of receptor-ligand interaction is considered as one of the fundamental approaches for rational drug design and so the prediction of such interactions by molecular docking has been gaining importance [17].

In the present study, the molecular docking study was done for some quinazoline-4-3H-one against human DHFR. This was followed by ADMET prediction and drug likeliness as well as drug score analysis of the docked compounds to evaluate the status of some quinazoline-4-(3H) -one as inhibitors of human DHFR.

MATERIALS AND METHODS

The study comprised of 27 compounds belonging to quinazoline-4-(3H)-one (Fig.1) along with one standard drug methotrexate. The selected compounds have different substituents as shown in Table 1. Molinspiration (http://www.molinspiration.com) and OSIRIS Property explorer (http://www.organic-chemistry.org/prog/peo/) were used to calculate logP, solubility, drug likeliness, polar surface area, molecular weight, number of atoms, number of rotatable bonds, volume, drug score and number of violations to Lipinski's rule. PreADMET (http://preadmet.bmdrc.org/) server was also used to test drug-likeliness and ADMET (Absorption, Distribution, Metabolism, Excretion and Toxicity) profile. The OSIRIS program was used to predict the overall toxicity of the most active derivatives (as it may reveal or indicate the presence of some fragments generally responsible for the irritant, mutagenic, tumorigenic, or reproductive effects of the tested compounds).

The physical properties of the ligands were determined. The similarity coefficient of the ligands was compared with the standard drug methotrexate and a cluster tree representing the similarity of the molecules was generated by ChemMine tools (http://chemmine.ucr.edu). Automated molecular docking was performed using the AutoDock 4.0 suite [18].

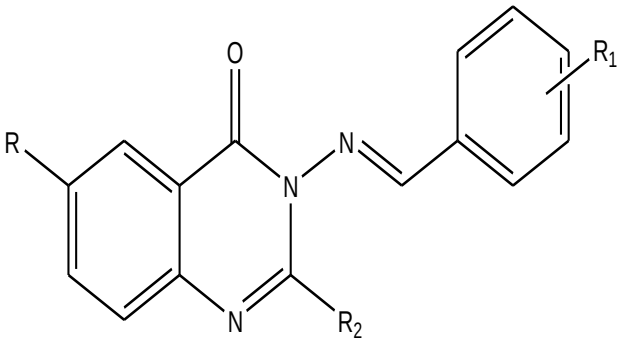

Fig. 1: Structure of 3-amino-2-aryl-4(3H)-quinazolinone

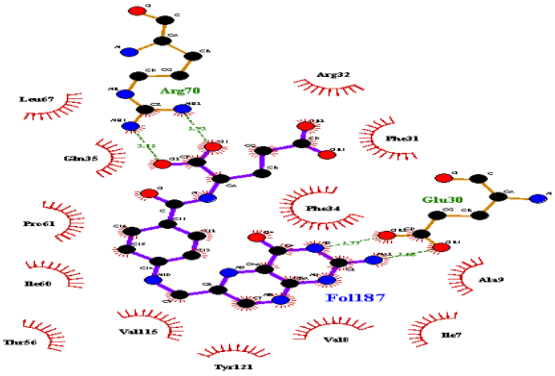

Fig. 2: LigPlot generated snapshot of the residues in the active site of 1DHF interacting with the natural ligand Folate.

Table 1: The substituents of the 3-amino-2-aryl-4(3H)-quinazolinone

Compounds	R	R_1	R_2
4a	H	3,5-Cl	Ph
4b	H	3-NO_2-4-Cl	Ph
4c	H	4-CF_3	Ph
4d	H	3-Cl	Ph
4e	H	2,3-Cl	Ph
4f	H	2,6-Cl	Ph
4g	H	3,4-F	Ph
4h	H	3-CF_3	Ph
4i	6-Br	2-F	Me
4j	6-Br	3-F	Me
4k	6-Br	4-F	Me
4l	6-Br	2-CF_3	Me
4m	6-Br	3-Cl	Me
4n	6-Br	2,4-Cl	Me
4o	6-Br	2,6-Cl	Me
4p	6-Br	3,4-F	Me
4q	6-Br	2-Cl, 5-NO_2	Me
4r	6-Br	4-Cl, 3-NO_2	Me
4s	6-Br	2-F, 3-CF_3	Me
4t	6-Br	3,4-OMe	Me
4u	6-Br	2,3-OMe	Me
4v	6-Br	2,5-OMe	Me
4w	6-Br	3-NO_2	Me
4x	6-Br	2-OH	Me
4y	6-Br	2,4-OMe	Me
4z	6-Br	5-Cl, 3-OH	Me
4	H	2,3-Cl	Me

The three dimensional structure (Fig 2) of the human dihydrofolate reductase was retrieved from the protein data bank (PDB ID: 1DHF) [19]. All water molecules and ligands were removed from the PDB file prior to docking. The receptor molecule was prepared by adding all missing hydrogen and side chain atoms, using the graphic user interface of AutoDock tools (ADT) [20]. The ligand files were also prepared from the 27 compounds used in this study, by adjusting the number of rotatable and non-rotatable bonds in the ligand molecules to assist in flexible docking process. The number of active torsions was set to the maximum number of atoms. As AutoDock requires pre-calculated grid maps, one for each atom type, present in the ligand being docked for storing the interaction potential energy, the grid was prepared in a way that it surrounded the active site based on the amino acid residues, which are involved in folate binding. The grid box size was set at 90, 90, and 90 A° (x, y, and z respectively) using AutoGrid 4.0 Program integrated in AutoDock 4.0. Twenty seven separate molecular docking experiments were set up using Lamarckian Genetic Algorithm (LGA) keeping all other parameters set in default mode. The top ranked model in the lowest energy cluster with maximum cluster size was considered for further interaction studies. Interaction has been compared based on the amino acid residues interacting with the natural ligand folate in the active site of hDHFR [Fig 2]. The docking result was converted from .dlg format to .pdb format by using python script. The compounds were structurally aligned to get a ligand based pharmacophore using PharmaGist tool [21].

RESULTS

Twenty-seven compounds used in this study have successfully qualified Lipinski's Rules, CMC like rule (except 4a and 4f), MDDR like rule and WDI like rule (Table 2). Ligands tested in this study were predicted to have good oral bioavailability (Table 3). Some of the compounds (4e, 4f and 4) have shown excellent permeability, while others have relatively less or poor (in some cases) permeability (Table 4). The physical properties like ionization potential, electronic energy and dipole plays an important role in activity of compounds (data not shown). The drug score and drug likeliness of the ligands were also predicted (Table 5). It revealed that drug score of compounds (4t, 4v, 4k, 4i, 4m, 4o and 4y) in the range of 0.5-0.66 and the rest of the compound in the range of 0.2 0.5.

Table 2: Data representing the qualification of the substituents for drug likeliness using CMC like rule, MDDR like rule and WDI like rule along with Rule of Five as predicted using OSIRIS server

Compound	CMC like rule	MDDR like rule	Rule of five	WDI like rule
4a	Not qualified	Mid structure	Suitable	90%
4b	Qualified	Mid structure	Suitable	90%
4c	Qualified	Mid structure	Suitable	90%
4d	Qualified	Mid structure	Suitable	90%
4e	Qualified	Mid structure	Suitable	90%
4f	Not qualified	Mid structure	Suitable	90%
4g	Qualified	Mid structure	Suitable	90%
4h	Qualified	Mid structure	Suitable	90%
4i	Qualified	Mid structure	Suitable	90%
4j	Qualified	Mid structure	Suitable	90%
4k	Qualified	Mid structure	Suitable	90%
4l	Qualified	Mid structure	Suitable	90%
4m	Qualified	Mid structure	Suitable	90%
4n	Qualified	Mid structure	Suitable	90%
4o	Qualified	Mid structure	Suitable	90%
4p	Qualified	Mid structure	Suitable	90%
4q	Qualified	Mid structure	Suitable	90%
4r	Qualified	Mid structure	Suitable	90%
4s	Qualified	Mid structure	Suitable	90%
4t	Qualified	Mid structure	Suitable	90%
4u	Qualified	Mid structure	Suitable	90%
4v	Qualified	Mid structure	Suitable	90%
4w	Qualified	Mid structure	Suitable	90%
4x	Qualified	Mid structure	Suitable	90%
4y	Qualified	Mid structure	Suitable	90%
4z	Qualified	Mid structure	Suitable	90%
4	Qualified	Mid structure	Suitable	90%

Table 3: Molecular descriptor properties of the ligands

Compound	miLogP	TPSA	nON	nOHNH	Nviolations	nrotb	volume	natoms
4a	5.887	47.261	4	0	1	3	321.379	27.0
4b	5.16	93.08	4	0	0	3	317.532	27.0
4c	4.25	47.261	4	0	0	3	288.64	25.0
4d	5.25	47.26	4	0	1	3	307.843	26.0
4e	5.863	47.261	4	0	1	3	321.379	27.0
4f	5.86	47.26	4	0	1	3	321.37	27.0
4g	4.85	47.26	4	0	0	3	304.17	27.0
4h	5.47	47.26	4	0	1	4	325.60	29.0
4i	3.47	47.26	4	0	0	2	262.27	22.0
4j	3.5	47.26	4	0	0	2	262.27	22.0
4k	3.52	47.26	4	0	0	2	262.27	22.0
4l	4.20	47.26	4	0	0	3	288.64	25.0
4m	4.01	47.26	4	0	0	2	270.88	22.0
4n	4.64	47.26	4	0	0	2	284.41	23.0
4o	4.62	47.26	4	0	0	2	284.41	23.0
4p	3.61	47.261	4	0	0	2	267.20	23.0
4q	3.92	93.08	7	0	0	3	294.21	25.0
4r	3.52	47.261	4	0	0	2	262.27	22.0
4s	4.32	47.261	4	0	0	3	293.57	26.0
4t	3.00	65.729	6	0	0	4	308.43	25.0
4u	3.18	65.729	6	0	0	4	308.43	25.0
4v	3.40	65.729	6	0	0	4	308.43	25.0
4w	3.2	93.08	7	0	0	3	280.67	24.0
4x	3.3	67.489	5	1	0	2	265.36	22.0
4y	3.40	65.729	6	0	0	4	308.43	25.0
4z	3.954	67.489	5	1	0	2	278.898	23.0
4	4.62	47.261	4	0	0	2	284.41	23.0

Table 4: preADME prediction of ligands

Compound name	HIA%	Caco-2 nm/sec	MDCK nm/sec	In vitro plasma%	In vitro blood barrier
4a	98.06	45.898	15.94	96.47	0.84
4b	99.142	17.55	0.044	93.484	0.027
4c	97.68	27.43	0.044	92.11	0.135
4d	97.84	42.28	44.058	93.244	2.07
4e	98.06	45.54	25.09	96.07	1.107
4f	98.06	44.77	34.26	95.998	2.05
4g	97.62	44.77	0.182	93.022	0.269
4h	97.668	27.45	0.044	93.718	0.127
4i	97.589	35.543	0.0958	96.408	2.31
4j	97.589	35.46	0.053	100	1.39
4k	97.589	35.455	0.046	99.18	0.996
4l	97.63	42.27	0.020	100	0.1945
4m	97.809	38.752	0.094	100	1.319
4n	98.003	42.6359	0.0412	100	0.79
4o	98.033	47.7122	0.125	100	1.38
4p	97.592	35.998	0.025	98.44	0.491
4q	99.14	17.55	0.023	100	0.292
4r	99.143	17.34	0.0208	100	0.201
4s	97.64	43.077	0.021	98.35	0.159
4t	97.485	37.517	0.024	95.31	0.241
4u	97.485	37.62	0.026	92.21	1.88
4v	97.485	37.62	0.028	92.71	1.93
4w	99.38	18.775	0.0323	100	0.194
4x	96.169	21.197	0.138	94.513	0.623
4y	97.48	37.06	0.028	89.708	0.358
4z	96.56	22.355	0.037	98.24	0.49
4	97.64	39.17	75.66	91.17	1.67

Table 5: Fragment based drug-likeliness of the ligands

Compound	cLogP	Solubility	MW	Drug likeness	Drug Score
4a	5.26	-6.27	393	1.71	0.23
4b	3.77	-5.44	404	5.07	0.45
4c	4.65	-5.44	393	5.19	0.4
4d	5.26	-5.54	359	5.51	0.31
4e	5.26	-6.27	394	5.59	0.4
4f	4.65	-6.27	394	6.1	0.31
4g	4.16	-5.43	361	2.68	0.42
4h	4.8	-5.58	393	-1.84	0.21
4i	3.42	-4.78	362	1.44	0.59
4j	3.42	-4.78	360	0.12	0.4
4k	3.42	-4.78	360	1.91	0.61
4l	4.12	-5.24	410	-6.99	0.27
4m	3.97	-5.2	376	2.72	0.55
4n	4.59	-5.2	411	3.17	0.44
4o	3.97	-5.94	411	3.76	0.56
4p	3.48	-5.09	378	0.19	0.47
4q	3.71	-5.84	421	1.5	0.45
4r	3.71	-5.84	428	3.16	0.49
4s	4.18	-5.56	402	-4.45	0.25
4t	3.15	-4.5	402	4.56	0.66
4u	3.15	-4.5	402	3.02	0.65
4v	3.15	-4.5	402	3.23	0.66
4w	3.09	-5.1	387	2.57	0.6
4x	3.06	-4.17	358	2.7	0.71
4y	3.15	-4.5	402	1.57	0.61
4z	3.67	-4.9	392	3.41	0.6
4	3.89	-5.1	332	4.94	0.49

The structural similarities of the compounds between each pair of molecules and also with the standard drug methotrexate were calculated. The cluster diagram revealing the relatedness amongst the molecules considering methotrexate as a reference has been shown in Fig. 3. The fate of a promising drug depends on its toxicity. The therapeutic index of a drug would be higher when it shows low toxicity/side effects. Based on this we have performed toxicity predication using Osiris Property Explorer. Results revealed that the compounds have low toxicity. The prediction using Osiris Property Explorer was shown in color codes. Green color represents low toxicity, yellow represents the mediocre toxicity, and red represents high toxicity as shown in table 6.

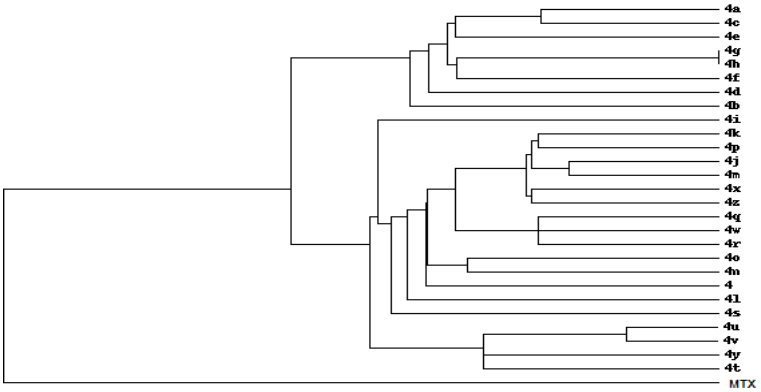

Fig. 3: **The cluster image showing the structural relationship of ligands (4, 4a-4z) with Methotrexate (MTX).The cluster tree drawn by ChemMine tools (http://chemmine.ucr.edu).**

Table 6: Toxicity prediction as per output of Orisis programme

Compound	Mutagenic	Tumorogenic	Irritant	Reproductive effect
4a	Green	Green	Green	Yellow
4b	Green	Green	Green	Yellow
4c	Green	Green	Green	Yellow
4d	Green	Green	Green	Yellow
4e	Green	Green	Green	Yellow
4f	Green	Green	Green	Yellow
4g	Green	Green	Green	Yellow
4h	Green	Green	Green	Yellow
4i	Green	Green	Green	Green
4j	Green	Green	Green	Yellow
4k	Green	Green	Green	Green
4l	Green	Green	Green	Green
4m	Green	Green	Green	Green
4n	Green	Green	Green	Green
4o	Green	Green	Green	Green
4p	Green	Green	Green	Green
4q	Green	Green	Green	Green
4r	Green	Green	Green	Green
4s	Green	Green	Green	Green
4t	Green	Green	Green	Green
4u	Green	Green	Green	Green
4v	Green	Green	Green	Green
4w	Green	Green	Green	Green
4x	Green	Green	Green	Green
4y	Green	Green	Green	Green
4z	Green	Green	Green	Green
4	Green	Green	Green	Green

The molecular docking results revealed that the docked complex of 27 compounds had less binding energy than methotrexate as shown in table 7 .The docking model of the ligand and hDHFR are shown in Fig. 4. The molecular alignment results have shown that all the compounds under study along with the standard drug methotrexate have occupied the same cavity (Fig 5) as is occupied by the natural ligand folate. All the 27 compounds were used to develop a ligand based pharmacophore (Fig. 6) using PharmaGist tool. Pharmacophore with a score of 66.813 showed the following characteristics: five spatial features out of which three are aromatic rings and two are hydrogen bond acceptors. There are no negative or positive centers, hydrophobic groups or hydrogen bond donors.

DISCUSSION

Molecular descriptor properties

The selected compounds used in this study were evaluated as potential hDHFR inhibitors. The oral bioavailability of the compounds projected as potential drugs were evaluated by determining the molecular weight, number of rotatable bonds (nrotb), number of hydrogen bonds (nON and nOHNH), and drug's polar surface (TPSA). Since the individual molecular weights of all the compounds were less than 500, the number of the rotatable bond were <10, the number of hydrogen bond donors and acceptors were < 12, and TPSA values being <140, they qualified to be an ideal oral drug. Ligands tested in this study were also predicted to have good oral bioavailability.

Calculation of the fragment based drug-likeliness of the compounds signifies that the compounds have the same fragments as compared to the existing drug. The drug-likeliness values of all the compounds are reasonably acceptable (except 4h,4i and 4s) as shown in Table 2. The higher drug-likeliness values are found in the case of compounds 4c, 4d, 4e and 4f. Results indicated that these four compounds have the most fragments similar to existing potent drugs to fulfill the potentiality of being drugs.

The drug score values [Table 3] were also calculated which took into account the effect of drug-likeliness, LogP, solubility, molecular weight, and toxicity risk together.

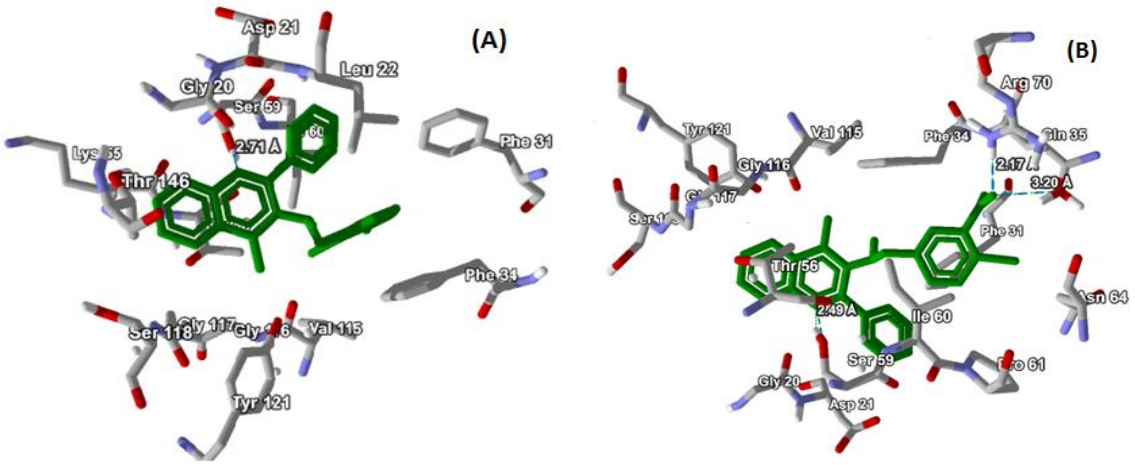

Fig. 4: (A) Ligand 4e docked with hDHFR; (B) ligand 4b docked with hDHFR.

Table 7: Binding energy and Inhibition constant of ligand -human DHFR interaction for each test compound

Compound	Binding energy Kcal/Mol	Inhibition constant (nM)
4a	-11.73	2.5
4b	-12.33	0.91
4c	-11.2	6.61
4d	-11.7	2.35
4e	-12.38	0.846
4f	-11.88	1.96
4g	-11.26	5.55
4h	-11.28	5.43
4i	-10.45	21.82
4j	-10.65	15.49
4k	-10.62	16.23
4l	-10.96	9.26
4m	-11.14	6.79
4n	-11.39	4.44
4o	-10.76	13.0
4p	-10.05	42.94
4q	-11.52	3.59
4r	-11.5	3.72
4s	-10.56	18.06
4t	-10.41	10.09
4u	-10.92	9.91
4v	-10.56	18.18
4w	-12.15	1.23
4x	-10.79	12.35
4y	-10.62	16.35
4z	-10.38	24.81
4	-10.47	21.25
MTX	-8.62.	479.78

Any compound that is considered to be a better drug candidate should exhibit better drug score. Our data showed that compound 4x has the best score (0.71), the compounds 4t,4v,4k,4i,4m,4o and 4y were in the range of 0.5-0.66, and the rest of the compounds were in the range of 0.2-0.5. The hydrophobicity of drugs could be inferred from LogP values [Table 3]. It was found that hydrophobicity and retention time of the drug inside the host are directly related i.e. higher the hydrophobicity, higher is the retention time of the drug in the body [23].

ADME prediction

In the modern drug designing process, computational approaches like preADMET prediction; MDCK and Caco-2 cell permeability, etc. serve as computational screening model for the prediction of intestinal drug absorption. All the compounds under study have qualified HIA%, *in vitro* plasma% (>90% in all the cases) and Caco-2 cell permeability (>25 nm/Sec) to be a good drug candidate. Some of the compounds have shown excellent permeability, while others have relatively less or poor (in some cases) permeability in relation to qualify as CNS drug and MDCK permeability as shown in Table 4. Less permeability is predicted because of the lesser solubility; and solubility, to a certain extent, depends on the arrangement of molecules in the crystal. It is to be noted that the topological aspects cannot be predicted via atom types or substructure fragments.

Molecular Docking and Pharmacophore study

The docked complexes of the receptor (original PDB structure (1DHF:A) and compounds in terms of the occupancy of the active site were compared. The molecular alignment results have shown that all the compounds under study along with the standard drug methotrexate have occupied the same cavity as is occupied by the natural ligand folate. The active site of the human dihydrofolate reductase (hDHFR) is represented by Ile-7, Ala-9, Trp-24, Glu-30, Gln-35, Asn-64, Arg-70, Val-115, Tyr-121 and Thr-136 [24]. It can be inferred that the compounds have an affinity for the active site and can act as competitive inhibitors to the natural ligand.

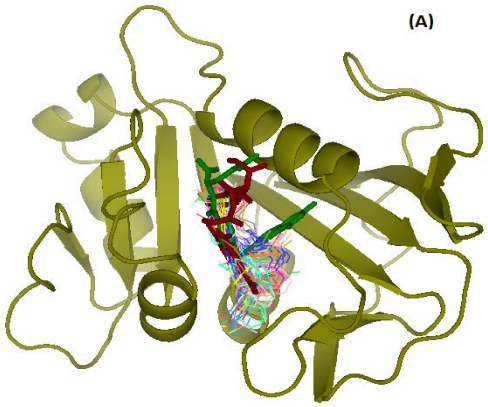

Fig. 5: (A) Docking model of ligands with hDHFR (PDB ID- 1DHF) protein (Folic Acid and methotrexate are represented in green and red sticks respectively, and the other 27 molecules are shown in line representation)

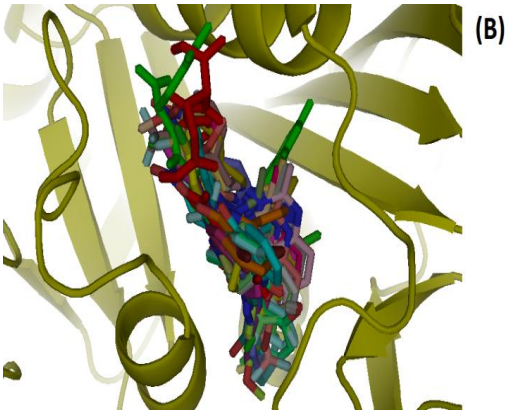

Fig. 5: (B) Zoomed view of the active site showing all the docked molecules (ligands are represented in different color sticks including folic acid and methotrexate represented in green and red sticks respectively)

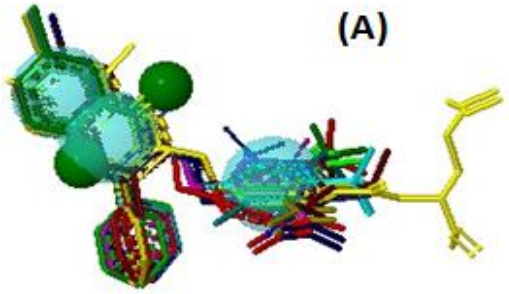

Fig. 6: (A) Structural alignment of 27 molecules along with the reference compound, methotrexate (methotrexate is shown in yellow color)

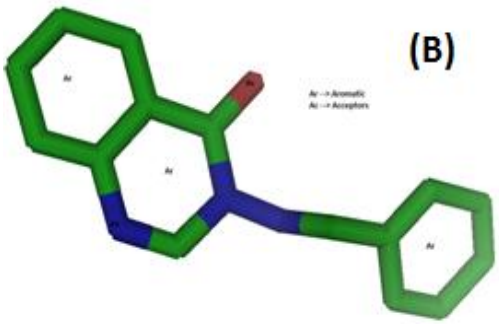

Fig. 6: (B) Structural representation of the derived Pharmacophore

The compounds were evaluated in terms of their binding mode to hDHFR. Based on the binding free energy (ΔG binding) of the protein-ligand interaction and inhibition constant (*Ki*), and one of the 10 models was chosen to be the best one. The docking result showed that all 27 compounds have low binding energy and inhibition constant as compared to the standard drug methotrexate. The minimum binding energy (maximum stability) was found in case of the compound 4e (-12.38 Kcal/Mol). The N1 of 4e forms hydrogen bond with the Ser-59 with a distance of 2.71Å. The amino acids Val-115, Phe-31, Phe-34, Tyr-121, Thr-136 and Asp-21 are found to be involved in making hydrophobic interactions with 4e. Interestingly, all these amino acids are also present in the active site of hDHFR, which infers that 4e binds to the active site region of the enzyme. The ligand 4b has also formed significant stable complex on docking. Similar to 4e, the N1 atom of 4b formed hydrogen bond with the Ser-59. It was reported that the tested quinazoline's recognition with the key amino acid Glu-30 and Ser-59 are essential for binding and biological activity [25]. The maximum binding energy was found in 4p (-10.05 Kcal/mol) which did not form hydrogen bond with the residues of the receptor. It was observed from the calculated binding energies that incorporation of phenyl at 2-C increased the interaction with the enzyme in comparison to compounds substituted with methyl at 2-C. The compounds 4a- 4h have binding energies in the range of -11.28 to -12.38 Kcal/Mol. The inhibition constant is directly proportional to the binding energy as shown in table 7. Many authors have used ligand-based approach for pharmacophore modeling of species-specific DHFR inhibitors. Moreover, a pharmacophore model for hDHFR (human) inhibitors has also been modeled [26]. All the 27 compounds were used to develop a ligand based pharmacophore [Fig. 6 (B)] using PharmaGist tool, which could be used further for the development of new, improved and optimized drug acting as inhibitor to hDHFR.

The pharmacophore has 3 aromatic rings and two hydrogen bond acceptors which enables in making several non covalent interactions like hydrophobic-hydrophobic interactions, hydrogen bonding, pi cloud interactions, etc.

This pharmacophore is qualifying all the four parameters of Lipinski's rule of five and thus could be considered as a lead molecule to generate new conformations for virtual screening library along with more modifications which could enhance its therapeutic index by enhancing the kind of interactions it could possibly make with the target protein.

CONCLUSIONS

Structure based drug design is significantly based on the protein-ligand interaction. The molecular docking study signified that the compounds can act as a putative inhibitor of hDHFR. The binding energy was found to be lesser than that of methotrexate. The compounds have also successfully qualified the rule of five, CMC like rule, WDI like rule and MDDR like rule. Every compound possessed apt pharmacological properties based on the results of Lipinski's Rule, hydrophobicity (based on log P value), and good drug likeliness and drug score. Moreover, the compounds have low toxicity value. The compounds were predicted to be safe (non-mutagenic as well as non-carcinogenic). This study has enabled to broaden the vision for the generation of more specific drugs for hDHFR, and may pave the way for the production and identification of more effective drugs.

Conflicts of Interest: All authors have none to declare.

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