ejpmr, 2020,7(7), 528-539

SJIF Impact Factor 6.222

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Research Article

ISSN 2394-3211

EJPMR

IN SILICO STUDIES OF IMIDAZOLE DERIVATIVES AS GLUCOSAMINE-6-PHOSPHATE SYNTHASE INHIBITORS

K. Hemalatha*, R. Kamali, M. Malarvizhi, S. Pragathi, M. Tamizhmathy and K. Girija

Department of Pharmaceutical Chemistry, College of Pharmacy, Mother Theresa Post Graduate and Research Institute of Health Sciences, (A Government of Puducherry Institution), Indira Nagar, Gorimedu, Puducherry-06.

***Corresponding Author: K. Hemalatha**
Department of Pharmaceutical Chemistry, College of Pharmacy, Mother Theresa Post Graduate and Research Institute of Health Sciences, (A Government of Puducherry Institution), Indira Nagar, Gorimedu, Puducherry-06.

Article Received on 26/04/2020 **Article Revised on 16/05/2020** **Article Accepted on 06/06/2020**

ABSTRACT

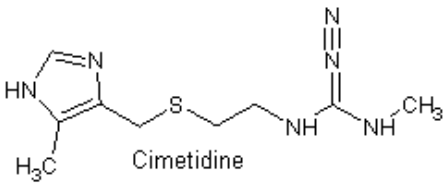
A series of imidazole derivatives has been designed. Absorption, distribution, metabolism and excretion (ADME) properties of all the designed derivatives have been evaluated *in-silico* using molinspiration and Swiss ADME tool to predict the physicochemical, pharmacokinetic, drug-likeness properties. None of the compounds violated Lipinski's rule of five properties and thus represented the possible use of the derivatives for developing compounds with drug-like properties. Molecular docking simulations were performed for the designed derivatives with Glucosamine-6- phosphate synthase (PDB ID: 2VF5). The binding energies of all the designed compounds have significant negative values as compared to the standard Ciprofloxacin and Fluconazole. These *in-silico* studies signified that the compounds can act as a putative inhibitor of glucosamine-6-phosphate synthase. Docking outcomes enhanced the designed hits act as promising antimicrobial agents.

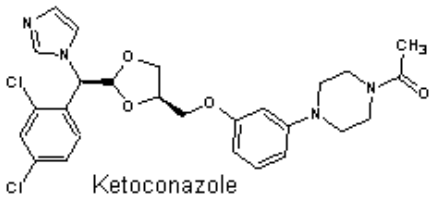
KEYWORDS: Imidazole, Molinspiration, Swiss ADME, Molecular docking, glucosamine-6-phosphate synthase.

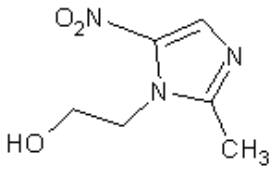
INTRODUCTION

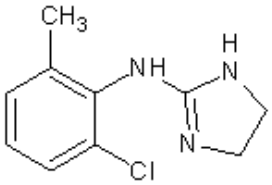
Increasing emergence of bacterial resistance to existing antibacterial has become a major concern among medicinal chemists around the world. So it has sparked keen interest in developing the new potent drugs with low toxicity and high bioavailability. An extensive use of antibacterial and their resistance has led to severe health problems in the hospitals and communities.[1] Imidazole, a member of azole heterocycles, having a five membered ring with 2 nitrogen atoms present at position 1 and 3 of the ring constitute an important pharmacophore. The imidazole scaffold is an interesting building block in various biomolecules such as Histidine, Histamine and Natural products i.e., Pilocarpine alkaloid (*Pilocarpus jaborandi*).[2,3] Being a polar and ionisable aromatic compound, it improves pharmacokinetic characteristics of lead molecules and thus is used as a remedy to optimize solubility and bioavailability parameters of proposed poorly soluble lead molecules.

Imidazole analogues has been considered as a versatile compounds due to their wide range of biological properties including Antimicrobial, Anti-inflammatory, Analgesic, Anti-ulcerative, Histamine H_3 antagonist, antioxidant, Antitumoral, Antiprotozoal, and Anti-diabetic activities.[4-13] Some imidazole derivatives such as Cimetidine, Etomidate, Ketoconazole, Metronidazole, Ornidazole, Azomycin, Oxiconazole, and Clonidine have found application in drug therapy.[14-15] D-fructose-6-phosphate amidotransferase, known under the trivial name of glucosamine-6-phosphate synthase (GlcN-6-P) which represents the effective target in antimicrobial chemotherapy. Glucosamine-6-phosphate synthase catalyzes the first step in hexosamine biosynthesis, converting D-fructose-6-phosphate (Fru-6-P) into D-glucosamine 6-phosphate (GlcN-6-P) using glutamine as the ammonia source and leading to the eventual formation of uridine 50-diphospho-N-acetyl-D-glucosamine (UDP-GlcNAc), the important point of metabolic control in the biosynthesis of amino sugar-containing macromolecules, which is necessary for the cell wall assembly in bacteria and fungi.[16] The present work involves designing of some imidazole derivatives (table 1) and study of its physicochemical properties, Molecular docking study against a bacterial target and evaluation of its *in silico* studies and ADMET properties.









MATERIALS AND METHODS

The study consisted of 12 compounds (Table 1) belonging to Imidazole Derivatives (Fig.1) along with standard. Molinspiration (http://www.molinspiration.com) and OSIRIS Property explorer (http://www.organicchemistry.org/prog/peo/) were used to calculate log P, solubility, drug likeness, polar surface area, molecular weight, number of atoms, number of rotatable bonds, volume, drug score and number of violations to Lipinski's rule. 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). All the designed derivatives were subjected to theoretical in-silico ADME prediction study Swiss ADME. physicochemical, pharmacokinetics, lipophilic and drug-likeness properties of all the derivatives based on various descriptors have been assessed.

Chemsketch was used to draw the 2D structure of the molecules to be analysed. The 2D structures of the designed derivatives have been converted to canonical SMILES format and used to compute the ADME properties in Swiss ADME tool.

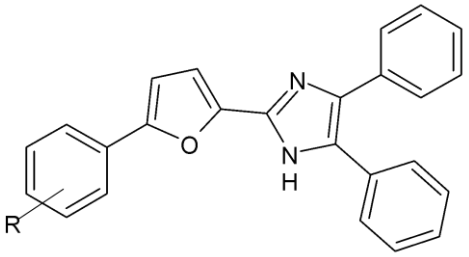


Fig1: Structure of Imidazole Derivative.

Molecular docking with Glucosamine-6-phosphate synthase

Protein Preparation

The crystallographic structure of Glucosamine-6-phosphate synthase in complex with glucosamine-6-phosphate (Figure 2) which were retrieved from the RCSB Protein Data Bank (PDB code: 2VF5) serves as docking receptor and the designed compounds are selected as ligand molecules. Before docking the screened ligands into the protein active site, the protein was prepared by deleting the substrate cofactor as well as the crystallographically observed water molecules and then protein was defined for generating the grid.

Ligand Preparation

ChemSketch, the chemically intelligent drawing interface freeware (http://www.acdlabs.com/download) was used to draw the structures of imidazole derivatives (Table 1), followed by generation of 3D structure in PDB format using Marvin sketch.

Automated docking was used to locate the appropriate binding orientations and conformations of various inhibitors into the 2VF5 binding pocket. To perform the task, the powerful genetic algorithm method implemented in the program AutoDock 4.0. was employed. Grid maps were generated by the AutoGrid program. Each grid was centered at the crystal structure of the corresponding 2VF5. Lamarckian Genetic Algorithm was employed as the docking algorithm. The grid dimensions were 60 Å X 60 Å X 60 Å with points separated by 0.375 Å. For all ligands, random starting positions, random orientations and torsions were used. During docking, grid parameters were specified for x, y and z axes as 38.808, 30.946 and 42.249 respectively. The Docking parameters Number of Genetic Algorithm (GA) runs: 25, Population size: 150, Maximum number of evaluation: 2,500,000, Maximum number of generation: 27,000 were used for this study. The structure with the lowest binding free energy and the most cluster members was chosen for the optimum docking conformation.

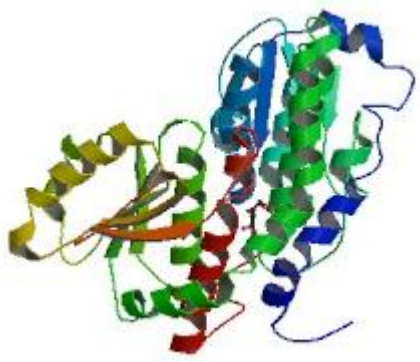
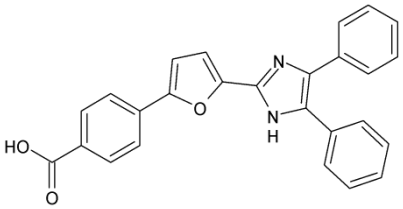
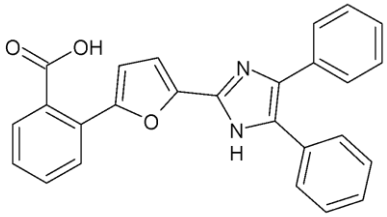
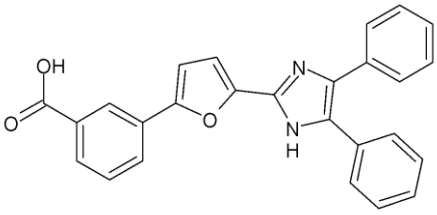
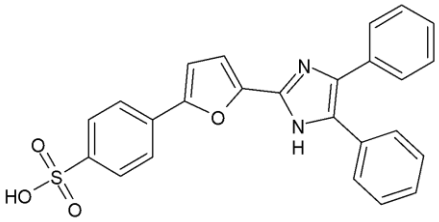
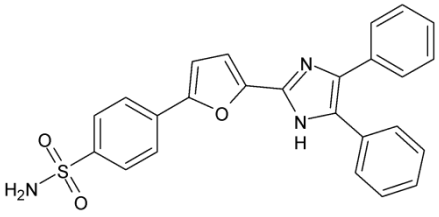
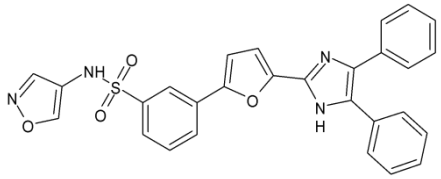
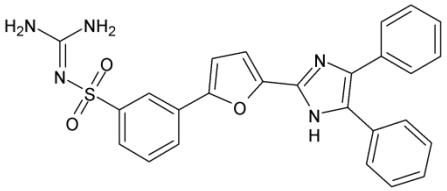
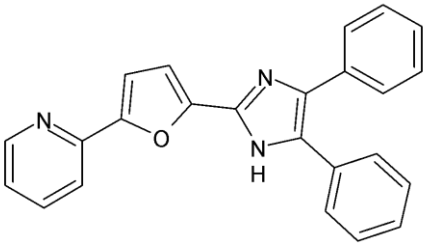
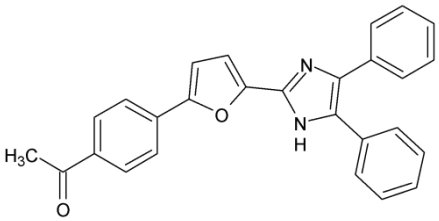
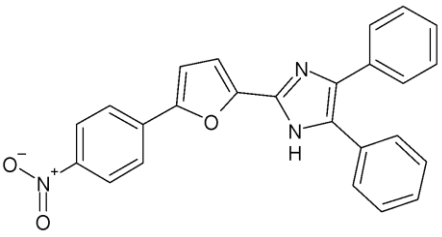
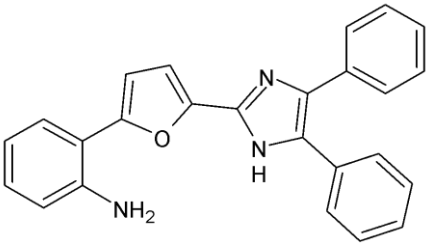
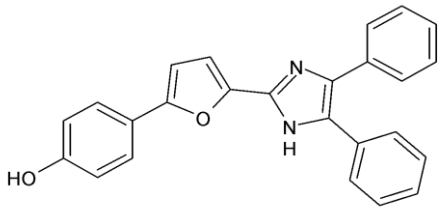
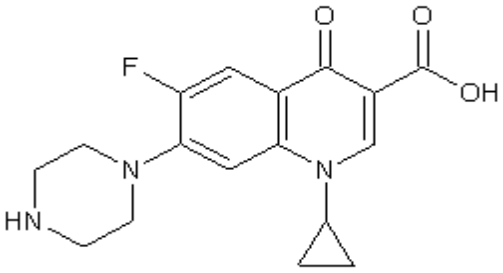
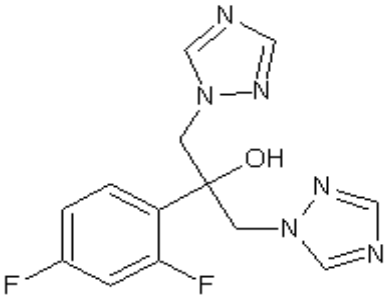


Figure 2: Crystal structure of Glucosamine-6-phosphate synthase (PDB ID: 2VF5)

Table 1: Compound code and its IUPAC Name.

S.No	Compound Code	Title Compounds
1	PABA-A_1	 4-[5-(4,5-Diphenyl-1H-imidazol-2-yl)furan-2-yl]benzoic acid
2	AA-A_2	 2-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]benzoic acid
3	MABA-A_3	 3-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]benzoic acid
4	SA-A_4	 4-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]benzene-1-sulfonic acid
5	SNA-A_5	 4-[5-(4,5-diphenyl-1H-imidazol-2-yl)furan-yl]benzene-1-

		sulfonamide
6	SMO-A_6	 4-[5-(4,5-diphenyl-1H-imidazol-2-yl)furan-2-yl]-(5-methyl-1,2-oxazol-3-yl)benzene-1-sulfonamide
7	SG-A_7	 2-({4-[5-{4,5-diphenyl-1H-imidazol-2yl-)furan-yl]benzene}sulfonyl)guanidine
8	2AP-A_8	 2-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]pyridine
9	4AA-A_9	 1-{4-[5-(4,5-diphenyl-1H-imidazol-2yl)furan-yl]phenyl}ethan-1-one
10	PNA-A_{10}	 2-[5-(4-nitrophenyl)furan-2-yl]-4,5-diphenyl-1*H*-imidazole
11	OPD-A_{12}	 2-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]aniline

12	PAP-A12	 4-[5-(4,5-diphenyl-1*H*-imidazol-2-yl)furan-2-yl]phenol
13	Ciprofloxacin	 1-cyclopropyl-6-fluoro-4-oxo-7-(piperazin-1-yl)-1,4-dihydroquinoline-3-carboxylic acid
14	Fluconazole	 2-(2,4-difluorophenyl)-1,3-bis(1H-1,2,4-triazol-1-yl)propan-2-ol

RESULTS AND DISCUSSION

Molecular properties calculations

Molecular properties, mainly hydrophobicity, molecular size, flexibility and the presence of various pharmacophoric features influence the pharmacokinetic and pharmacodynamics behaviour of molecules in the living organism, including bioavailability. So in order to achieve good bioavailable drugs, we have subjected a series of aryl imidazole derivatives (A1-A12) for the prediction of some basic pharmacokinetic properties under Lipinski's rule of Five.

Lipophilicity

All the compounds were subjected to computational study in order to filter the drugs for biological screening. for good membrane permeability logP value should be≤ 5.[17] All the title compounds were found to have log p values in the range of 3.44 to 6.36.

Absorption, Polar Surface area and Rule of Five Properties

High oral bioavailability is an important factor for the development of bioactive molecules as therapeutic agents. Good intestinal absorption, reduced molecular flexibility (measured by the number of rotatable bonds), low polar surface area or total hydrogen bond count (sum of donors and acceptors) are important predictors of good oral bioavailability. Molecular properties such as membrane permeability and bioavailability is always associated with some basic molecular descriptors with some basic molecular descriptors such as log P (partition coefficient), molecular weight (MW), or hydrogen bond acceptors and donors counts in a molecule.[18] Lipinski[19] used these molecular properties in formulating his ‘‘Rule of Five”. The rule states that most molecules with good membrane permeability have logP ≤ 5, molecular weight ≤500, number of hydrogen bond acceptors ≤10, and number of hydrogen bond donors ≤ 5.

Polar surface area (PSA) was determined by the fragment-based method of Ertl and coworkers.[20-21] A poor permeation or absorption is more likely when there are more than 5 H bond donors, 10 H-bond acceptors. Hydrogen-bonding capacity has been also identified as an important parameter for describing drug permeability.[22] The series of screened compounds have hydrogen bond donor and acceptors in considerable range as shown in Table 2.

Number of rotatable bonds is important for conformational changes of molecules under study and ultimately for the binding of receptors or channels. It is

revealed that for passing oral bioavailability criteria the number of rotatable bonds should be ≤10. The compounds in this series (A1-A12) possess a lower range of 'number of rotatable bonds' and therefore exhibit low conformational flexibility.

Molecular polar surface area (TPSA) is a very useful parameter for the prediction of drug transport properties. TPSA is a sum of surfaces of polar atoms (usually oxygen, nitrogen and attached hydrogen) in a molecule. All the title compounds (A1-A12) followed Lipinski's Rule of Five. The pharmacokinetic parameters were calculated online from Molinspiration Chemoinformatics.

Table 2: Pharmacokinetic properties important for good oral bioavailability.

Compound code	logp	TPSA	MW (gms)	nOH	nOHNH	nrotb	volume
PABA-A_1	6.36	79.12	406.44	5	2	5	359.08
AA-A_2	5.97	79.12	406.44	5	2	5	359.08
MABA-A_3	4.33	79.12	406.44	5	2	5	359.08
SA-A_4	3.44	96.19	442.50	6	2	5	371.52
SNA-A_5	5.14	101.99	441.51	6	3	5	374.80
SMO-A_6	4.04	114.02	522.59	8	2	7	441.29
SG-A_7	4.46	140.38	483.55	8	5	6	409.34
2AP-A_8	5.11	54.72	363.42	4	1	4	327.92
4AA-A_9	4.15	58.89	404.47	4	1	5	367.62
PNA-A_{10}	6.21	87.65	407.43	6	1	5	355.41
OPD-A_{11}	5.69	67.85	377.45	4	3	4	343.36
PAP-A_{12}	5.77	62.05	378.43	4	2	4	340.09
Ciprofloxacin	0.70	74.57	331.35	6	2	3	285.46
Fluconazole	-0.12	81.66	306.28	7	1	5	248.96

logP- Partition coefficient, TPSA-Topological polar surface area, MW- molecular weight, nOH- number of hydrogen bond acceptors, nOHNH- number of hydrogen bond donors, nrotb- number of rotatable bonds.

Predicted bioactivity score (Molinspiration calculations)

The molinspiration bioactivity prediction has to be removed because it has repeated again (http://www.molinspiration.com/docu/miscreen/druglikeness.html) is fast (100 000 molecules may be screened in about 30 min) and therefore allows processing of very large molecular libraries. Validation tests performed on various target classes (including of GPCR ligand, ion channel modulator, nuclear receptor ligand, kinase inhibitor, protease inhibitor, enzyme inhibitor) show 10–20-fold increases in hit rate in comparison with standard/random selection of molecules for screening. Calculated drug likeness scores of each compound were compared with the specific activity of other compounds and the results were compared with standard drugs. For organic molecules, the probability is if the bioactivity score is (40), then it is active, if (5.0 to 0.0) then moderately active, if (55.0) then inactive. The calculated value of the predicted bioactivity for compounds is given in Table 3.

Table 3: *In* silico predicted bioactivity of the designed compounds compared with standard drugs.

Compound Code	GPCR Ligand	Ion Channel Modulator	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor	Enzyme inhibitor
PABA-A_1	0.12	-0.04	0.20	0.01	-0.35	0.20
AA-A_2	0.18	-0.08	0.18	0.03	-0.33	0.22
MABA-A_3	0.14	-0.03	0.23	0.05	-0.33	0.22
SA-A_4	0.19	0.04	0.15	-0.30	-0.21	0.27
SNA-A_5	0.03	-0.10	0.22	-0.28	-0.23	0.25
SMO-A_6	0.09	-0.18	0.06	-0.28	-0.12	0.18
SG-A_7	0.10	-0.08	-0.03	-0.36	-0.14	0.12
2AP-A_8	0.23	0.06	0.38	-0.14	-0.39	0.29
4AA-A_9	0.05	-0.10	0.11	-0.17	-0.44	0.10
PNA-A_{10}	-0.01	-0.07	0.13	-0.20	-0.50	0.06
OPD-A_{11}	0.10	-0.04	0.26	-0.29	-0.33	0.18
PAP-A_{12}	0.16	0.01	0.30	0.01	-0.39	0.22
Ciprofloxacin	0.12	-0.04	-0.07	-0.19	-0.20	0.28
Fluconazole	0.04	0.01	-0.09	-0.23	-0.09	0.03

OSIRIS Calculations

Structure based drug design is now very routine work as many drugs fail to reach clinical phases because of ADME/TOX problems encountered. Therefore prediction of these problems before synthesis is a rational approach to minimize cost production of expensive chemicals. The Osiris calculations are tabulated in Table 4. Toxicity risks (mutagenicity, tumorigenicity, irritation, reproduction) and physicochemical properties (cLogP, solubility, drug likeness and drug score) of compounds (A1- A12) were calculated by the methodology developed by Osiris.[23] The toxicity risk predictor locates fragments within a molecule, which indicate a potential toxicity risk. Toxicity risk alerts are an indication that the drawn structure may be harmful concerning the risk category specified. The logP value of a compound, which is the logarithm of its partition coefficient between n-octanol and water, is a well-established measurement of the compound's hydrophilicity. Low hydrophilicities and therefore high logP values may cause poor absorption or permeation. It has been shown that for compounds to have a reasonable probability of good absorption, their clogP value must not be greater than 5.0. On this basis, all the compounds, except A9 and A12 possessed clogP values in the acceptable range.

Aqueous solubility

The aqueous solubility of a compound significantly affects its absorption and distribution characteristics. In general a low solubility goes along with a poor absorption and therefore the general aim is to avoid poorly soluble compounds. Our estimated logS value is a unit stripped logarithm (base 10) of a compound's solubility measured in mol/liter. There are more than 80% of the drugs on the market have a (estimated) logS value greater than -4. In the present series the values of logS are around -5. Further, Table-4 shows drug likeness of compounds (A1-A12) which is in the acceptable zone to be drug-like when compared with standard drugs. We have calculated overall drug score (DS) for the compounds A1-A12 and compared with that of standard drug ciprofloxacin and Fluconazole. Most of the designed derivatives are free form mutagenic, tumorigenic, irritation and reproductive toxicity.

Table 4: Osiris Calculation for bioavailability and Toxicity prediction.

Compounds	Solubility	clogP	Drug likeness	Drug score	Mutagenic	Tumorigenic	Irritation	Reproductive effect
A_1	-7.27	5.25	-1.79	0.16	Green	Green	Orange	Green
A_2	-7.27	5.25	0.12	0.13	Green	Green	Orange	Red
A_3	-7.27	5.25	-0.42	0.19	Green	Green	Orange	Green
A_4	-5.87	3.24	2.23	0.22	Green	Green	Green	Green
A_5	-7.16	4.7	4.44	0.29	Green	Green	Green	Green
A_6	-7.16	4.7	4.44	0.29	Green	Green	Orange	Green
A_7	-7.53	4.16	4.32	0.28	Green	Green	Orange	Green
A_8	-6.16	5.03	2.72	0.32	Green	Green	Orange	Green
A_9	-7.94	5.81	0.31	0.19	Green	Green	Orange	Green
A_{10}	-7.72	4.85	-7.34	0.32	Green	Green	Orange	Green
A_{11}	-7.34	5.26	0.36	0.22	Green	Green	Orange	Green
A_{12}	-6.96	5.57	3.1	0.26	Green	Green	Orange	Green
Ciproflox	-3.32	1.53	2.07	0.82	Green	Green	Green	Green
Fluconazole	-2.17	0.11	1.99	0.87	Green	Green	Green	Green

Green: Low risk, Orange: Medium risk, Red: High risk

***In silico* ADME Screening**

The pharmacokinetics and drug likeness prediction of compounds were performed online on Swiss ADME tool. Swiss ADME tool was used for online pharmacokinetic properties evaluation of compounds.[24] The online prediction was done to check the compound were inhibitors of cytochrome P450.In addition to the pharmacokinetic properties such as Gastrointestinal absorption, Blood-Brain Barrier penetration, Skin Permeation, synthetic associability and drug-likeness prediction like Lipinski, Ghose and Veber rules and bioavailability score.[25] The results were shown in table 5 and 6.

Table 5: Lipophilicity and Drug likeness properties of synthesized compounds.

S.No.	Compound code	Lipophilicity					DrugLikeness No. of Violations				
		I Log P	X Log P	W Log P	M Log P	Cons. Log P	L	G	V	E	M
1.	PABA-A_1	2.94	5.34	6.37	3.28	4.77	0	1	0	1	1
2.	AA-A_2	2.59	5.34	6.37	3.28	4.70	0	1	0	1	1
3.	MABA-A_3	3.79	4.92	6.01	3.10	4.84	0	1	0	1	0
4.	SA-A_4	2.48	4.56	7.00	3.0	4.37	0	1	0	1	0
5.	SNA-A_5	2.17	4.37	6.40	2.20	3.94	0	1	0	1	0
6.	SMO-A_6	0.00	4.78	7.19	2.50	3.77	1	3	0	1	0
7.	SG-A_7	2.89	3.86	5.71	2.37	3.76	0	3	1	1	0
8.	2AP-A_8	3.45	4.78	6.07	2.69	4.58	0	1	0	1	0
9.	4AA-A_9	3.75	5.49	6.87	3.23	5.26	0	1	0	1	1
10.	PNA-A_{10}	3.35	5.64	7.10	3.48	4.86	0	1	0	1	1
11.	OPD-A_{11}	3.45	5.13	6.26	3.16	4.76	0	1	0	1	1
12.	PAP-A_{12}	3.34	5.45	6.38	3.16	4.87	0	1	0	1	1
13.	Ciprofloxacin	2.24	-1.08	1.18	1.28	1.10	0	0	0	0	0
14.	Fluconazole	0.41	0.35	1.47	1.47	0.88	0	0	0	0	0

Cons. LogP- Consensus Log P, L-Lipinski, G-Ghose, V-Veber, E-Egan, M-Muegge rule, BAS- Bioavailability Score.

Table 6: Pharmacokinetics of compound: GI-Gastrointestinal absorption, BBB-Blood Brain Barrier penetration, P-gp- Substrate of the P-gp protein, CYP: Cytochrome P450, Log Kp-Skin Permeation Coefficient.

Compound code	GI	BBB	P-gp	CYP1A2	CYP2D6	$LogK_p$ (cm/s)	Bioavailability score
PABA-A_1	High	No	Yes	No	Yes	-4.99	0.55
AA-A_2	High	No	Yes	No	Yes	-4.99	0.55
MABA-A_3	High	No	Yes	Yes	Yes	-5.20	0.55
SA-A_4	Low	No	No	No	Yes	-5.76	0.55
SNA-A_5	Low	No	No	No	Yes	-5.89	0.55
SMO-A_6	Low	No	Yes	Yes	Yes	-6.70	0.55
SG-A_7	Low	No	No	No	No	-6.51	0.55
2AP-A_8	High	No	Yes	Yes	Yes	-5.12	0.55
4AA-A_9	Low	No	Yes	Yes	No	-4.87	0.55
PNA-A_{10}	Low	No	No	No	No	-4.87	0.55
OPD-A_{11}	High	No	Yes	Yes	Yes	-4.96	0.55
PAP-A_{12}	High	No	Yes	Yes	Yes	-4.74	0.55
Ciprofloxacin	High	No	Yes	No	No	-9.09	0.55
Fluconazole	High	No	No	No	No	-7.92	0.55

Molecular Docking study

Molecular docking studies of twelve imidazole derivatives and along with two standard drugs (Ciprofloxacin and Fluconazole) were carried out with the protein target Glucosamine-6-phosphate synthase using Autodock 4.0 to identify the binding mode of ligands and the intermolecular interaction between the ligands and the target protein. The docked pose for each of the compounds were evaluated and the pose with the lowest binding energy and the inhibition constant was thereby chosen as a hit molecule.

The lowest binding energy (best docking score) and inhibition constant indicated the highest ligand-protein affinity. The predicted binding energy observed for ligands SMO-A6, MABA-A3, SA-A4 and 4AA-A9 were found to be-8.72, -7.79, -7.55 and -7.00 kcal/mol respectively compared to the standard Ciprofloxacin (-5.69 kcal/mol) and Fluconazole (-5.18 kcal/mol). The inhibition constant value for the ligand SMO-A6, MABA-A3, SA-A4 and 4AA-A9 were found to be 407.08 nm, 1.94 µm, 2.94 µm and 7.37 µm respectively. (Table 7 and 8)

Compound SMO-A6 was found to establish two hydrogen bonds with Gln 348 and Lys 603 with a distance of 1.944 and 1.974 A° respectively and hydrophobic interactions with Lys487, Leu484, Val399, Asn305, Ala400, Ser401, Ser347, Thr, Ser34, Tyr304. similarly compound MABA-A3 formed three hydrogen bonds with Arg 311, Arg 311, Tyr 476 with the distance of 1.936, 1.686 and 1.735° respectively and hydrophobic interactions with Met308, Tyr312, Tyr304, Asn305, Ala496, Leu480. The docking pose of compound SMO-A6, MABA-A3, SA-A4, 4AA-A9 and standard are shown in figure 3, 4 and 5.

Table 7: Molecular interactions of designed compounds in the active site of Glucosamine-6-phosphate synthase (2VF5).

Compound code	No. of Hydrogen bonds formed	Amino acid involved in hydrogen bond interaction	Distance between donor and acceptor (A°)	Amino acid involved in Vander waals interactions
PABA-A_1	2	GLN348(O) Lys603(O)	2.022 1.863	Glu396, Val399, Ser401, Gly301, Tyr304, Ile326, Leu484
AA-A_2	1	Tyr476(O)	1.857	Tyr304, Gly301, Lys487, Leu484, Ala483, Leu480, Met306,Ala 496,Lys603
MABA-A_3	3	Arg311(O) Arg311(O) Tyr476(O)	1.936 1.686 1.735	Met308, Tyr312, Tyr304, Asn305, Ala496, Leu480
SA-A_4	2	Lys487(O) Tyr476(N)	1.756 1.872	Ala496, Ala483, Leu484, Leu480, Tyr304,Met308, Arg311
SNA-A_5	2	Lys603(O) Val399(H)	2.083 1.798	Tyr304, Thr302, Gly301, Leu480, Glu488, Leu484, Ala483, Lys487, Ala496
SMO-A_6	2	Gln348(N) Lys603(N)	1.944 1.974	Lys487, Leu484, Val399, Asn305, Ala400, Ser401, Ser347, Thr, Ser34, Tyr304,
SG-A_7	1	Ser303(O)	2.113	Gly301, Ser401, Glu488, Leu484, Ala483, Lys487, Ala496, Ile326, Tyr304, Ser303, Thr302
2AP-A_8	2	Tyr476(N) Tyr304(0)	2.012 Invisible	Met308, Asn306, Gly301, Leu484, Leu480, Ala483, Ala486, Lys487,
4AA-A_9	1	Val399(H)	Invisible	Lys603, Ser401, Ala400, Val399, Glu488, Leu484, Gly301, Thr302, Tyr304, Ile326, Lys487
PNA-A_{10}	0	------	------	Ile326, Tyr304, Gly301, Lys487, Leu484, Val399, Glu396, Lys608, Gln, Ser401,
OPD-A_{11}	1	Tyr476(O)	2.101	Ala496, Leu480, Gly301, Asn305, Met308, Tyr304, Ile326
PAP-A_{12}	3	Arg311(O) Tyr304(H) Tyr476(O)	2.196 2.162 Invisible	Ala496, Leu480, Met308, Tyr312
Ciprofloxacin	1	Lys487(O)	2.161	Tyr304, Met308, Asn305, Leu480, Tyr476, Ala483, Leu484
Fluconazole	1	Lys487(N)	1.884	Tyr304, Asn305, Gly301, Ala496, Ala483, Leu484, Glu495

Table 8: Molecular Docking reports for compounds A1-A12 along with standard drug against Protein 2VF5.

Compound code	Binding energy (kcal/mol)	Inhibition constant	Intermolecular energy (kcal/mol)	No. of conformations (10)	Rank
PABA-A_1	-5.61	77.33µm	-7.40	5	11
AA-A_2	-5.36	119.6µm	-6.86	6	8
MABA-A_3	**-7.79**	**1.94µm**	**-9.58**	**3**	**2**
SA-A_4	**-7.55**	**2.94 µm**	**-9.34**	**10**	**3**
SNA-A_5	-6.62	14.04 µm	-8.41	7	7
SMO-A_6	**-8.72**	**407.08 n**m	**-10.81**	**5**	**1**
SG-A_7	**-6.98**	**7.59 µm**	**-8.77**	**9**	5
2AP-A_8	-6.96	7.94µm	-7.85	8	6
4AA-A_9	**-7.00**	**7.37µm**	**-8.19**	**6**	**4**
PNA-A_{10}	-6.08	34.81 µm	-7.57	7	10
OPD-A_{11}	-6.16	30.34µm	-7.66	8	9
PAP-A_{12}	-6.16	30.31 µm	-7.66	9	9
Ciprofloxacin	-5.69	67.05 µm	-6.59	6	-
Fluconazole	-5.18	159.45 µm	-6.08	3	-

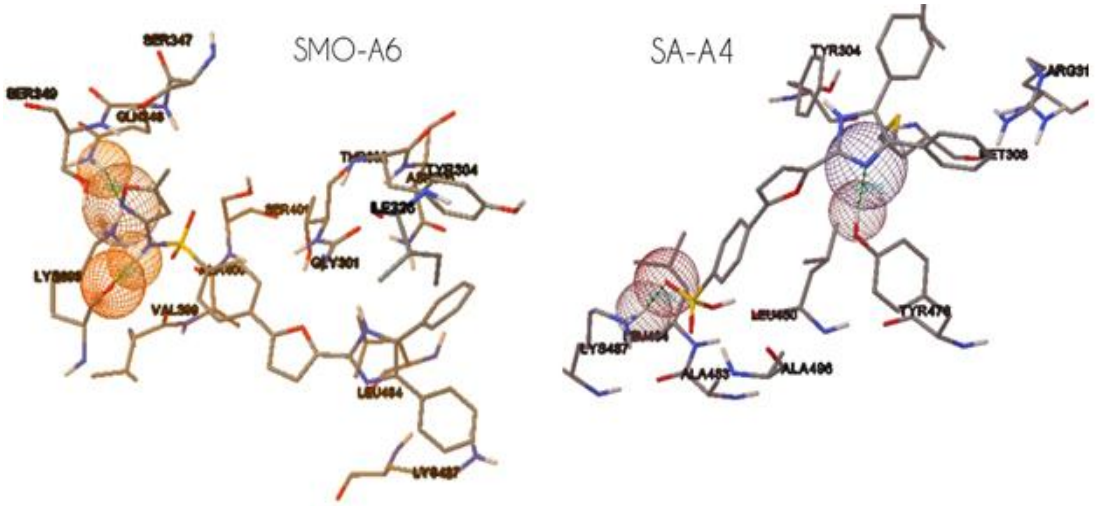


Fig 3: Docking pose of compound SMO-A6 and SA-A4 in the active site of the protein Glucosamine-6-phosphate synthase (2VF5).

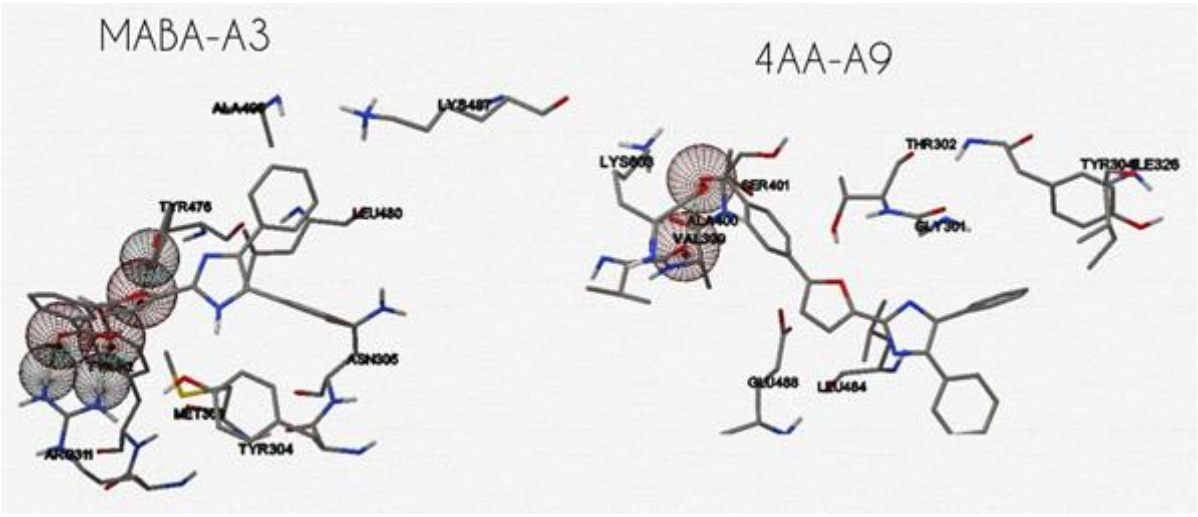


Fig 4: Docking pose of compound MABA-A3 and 4AA-A9 in the active site of the protein Glucosamine-6-phosphate synthase (2VF5).

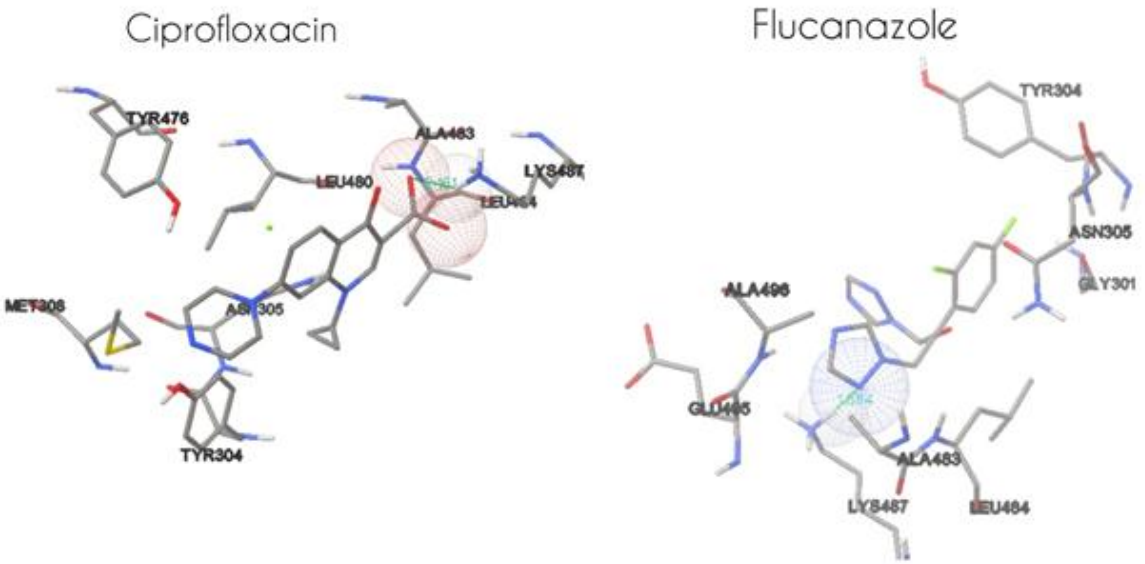


Fig 5: Docking pose of Standard Ciprofloxacin and Fluconazole in the active site of the protein Glucosamine-6-phosphate synthase (2VF5).

CONCLUSION

A series of 12 imidazole derivatives were designed and subjected to in-silico assessment using Molinspiration and Swiss ADME tool. All the designed compounds were according to lipinski's rule of five and showed to have a promising oral bioavailability. OSIRIS calculations revealed that all compounds were free from toxicities like mutagenic, tumorigenic, irritant and reproductive effects. Most of the docked compounds were found to possess good interaction in the receptor sites and exhibited bonding and non-bonding interactions with the active site residues of the receptor Glucosamine-6-phosphate synthase. Compounds SMO-A6, MABA-A3, SA-A4 and 4AA-A9 showed stronger binding affinity (least binding energy) compared to the standard drug and could act as a potent inhibitor against the organism. On the other hand, a promising ADME drug like profile for the present compounds offers the possibility of exepident additional modifications that could give rise to lead structures with enhanced inhibitory activity towards the drug receptor target.

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