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Structure Based Virtual Screening, Design, Synthesis and Biological Evaluation of Imidazoles as Anti-Diabetic Agents

K. Sreelatha*,Syeda Advia Sanobar, K. Sruthi, M. Sumakanth
Department of Pharmaceutical Chemistry,
RBVRR Women's College of Pharmacy, Barkatpura, Hyderabad,India.
3-4-343, Barkathpura, Lingampalli, Hyderabad, Telangana 500027.

Abstract

Virtual screening is a promising method for obtaining novel hit compounds in drug discovery. In the present study, SBVS is performed for imidazole using CHEMBLE database for different activities as antifungal, anticancer, antidiabetic, antibacterial.Among all screened derivatives, imidazoloneswere reported to have antidiabetic activity against SUR-1 receptors And also theseimidazoloneswere simultaneouslysubjected to virtualscreening PPARɣ receptors (as they are structurally similar to imidazolones) and were designed to emerge them as synergistic inhibitors of SUR-1 & PPARɣ for their possible antidiabetic activity.Then the HITS were identified and basic pharmacophoric features responsible for the activity were predicted. By keeping in view of these novel imidazolone derivatives were designed, docked and synthesized. The Synthesized compounds were characterized by physical and spectral data.

Keywords: Imidazoles, *Insilico* studies, Structure-based Virtual Screening,peroxisome proliferator-activated receptors,Sulfonylureas, Molecular Docking, characterization, Antidiabetic evaluation.

INTRODUCTION

Imidazoles are probably the most well known heterocycle which is common and important feature of a variety of natural products and medicinal agents[1].The imidazole nucleus is an important synthetic strategy in drug discovery[2].The imidazole is a planar, five membered heteroaromatic molecule with 3C and 2N atoms in 1 and 3 positions,this compound was earlier also called as glyoxaline as it was first prepared in (1858) from glyoxal and ammonia[3]. Derivatives of imidazole were reported for anti-cancer[4],anti-coagulants[5], anti-inflammatory[6], anti-bacterial[7], anti-fungal[8], anti-viral[9], anti-tubercular[10], anti-diabetic[11] and anti-malarial[12]analgesic[13] , anti-convulsant[14]activities.Diabetes mellitus is a complex metabolic disorder characterized by hyperglycemia due to inadequacy of insulin secretion and/ or insulin action[15]. The major symptoms include polydypsia, polyphagia, polyuria, blurred vision and weight loss. The world wide prevalence of diabetes among adults over 18 years of age has risen from 4.7% in 1980 to 8.5% in 2014.most commonly used treatment for type 2 diabetes include treatment with metformin, sulfonylureas, meglitinides, thiazoledinediones, DPP-4 inhibitors, SGLT-2 inhibitors and insulin therapy.SUR-1 and PPAR agonists are used increasingly to counteract the effects of diabetes[16].

Sulfonylureas stimulate insulin secretion from pancreatic β-cells and are widely used to treat type 2 diabetes[17]. Their principal target is the ATP-sensitive potassium (KATP) channel[18], which plays a major role in controlling the β-cell membrane potential. Inhibition of KATP channels by glucose or sulfonylureas causes depolarization of the β-cell membrane; in turn, this triggers the opening of voltage-gated $Ca2^{+}$channels[19], eliciting $Ca2^{+}$ influx and a rise in intracellular $Ca2^{+}$ which stimulates the exocytosis of insulin-containing secretory granules[20,21]. KATP channels are also found at high density in a variety of other cell types, including cardiac, smooth, and skeletal muscle, and some brain neurons[22], They primarily act by binding to the SUR subunit of the ATP-sensitive potassium (KATP) channel and inducing channel closure. SUR-1 is an ATP-binding cassette (ABC) transporter with a novel function[23]. In contrast to other ABC proteins, it serves as the regulatory subunit of an ion channel. The ATP-sensitive (K_{ATP}) channel is an octameric complex of four pore-forming Kir6.2 subunits and four regulatory SUR1 subunits[24-25], and it links cell metabolism to electrical activity in many cell types. ATPase activity at the nucleotide-binding domains of SUR results in an increase in K_{ATP} channel open probability. Conversely, ATP binding to Kir6.2 closes the channel. Metabolic regulation is achieved by the balance between these two opposing effects[26].

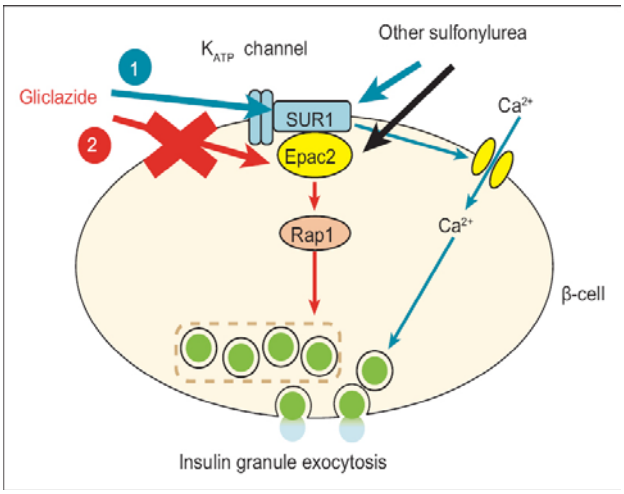

Figure 2: Effect of Sulfonyl ureas inhibitor on potentiation of insulin secretion

PPAR agonists are drugs which act on the peroxisome proliferator-activated receptor. They are used for the treatment of the metabolic syndrome, diabetes mellitus, mainly for lowering triglycerides and blood sugar[27,28]. There are four classes of PPAR agonists (alpha, gamma, delta, pan and dual). PPARγ are members of the nuclear hormone receptor family of ligand-activated transcription factor[29].They play a key role in regulating the insulin sensitivity, adipocyte differentiation, inflammation and cell growth[30]. PPAR gamma is the main target of thiazolidinediones used in diabetes mellitus characterised by insulin resistance[31].

Thiazolidinediones, acting via PPARγ, influence free fatty acid flux and thus reduce insulin resistance and blood glucose levels[32]. PPARγ agonists are therefore used to treat type 2 diabetes.Glycemic mechanism of action of thiazolidinedione "insulin sensitizers" (using an adipocyte for illustration purposes)[33]. These drugs are synthetic ligands for the transcription factor PPARγ, a member of a superfamily of nuclear receptors including thyroid and steroid receptors[34]. PPARγ is expressed in multiple tissue types (e.g. skeletal muscle, fat & liver). PPARγ stimulation upregulates the expression of genes involved in lipid & glucose metabolism[35], insulin signal transduction[36], and adipocyte differentiation. As illustrated, one mechanism contributing to the hypoglycemic effect of thiazolidinediones is an increased expression of the glucose transporter GLUT4. The increased expression of GLUT4 (in addition to mediators of insulin signal transduction) increases the ability of cells (e.g. adipocytes) to take up glucose when stimulated by insulin[37].

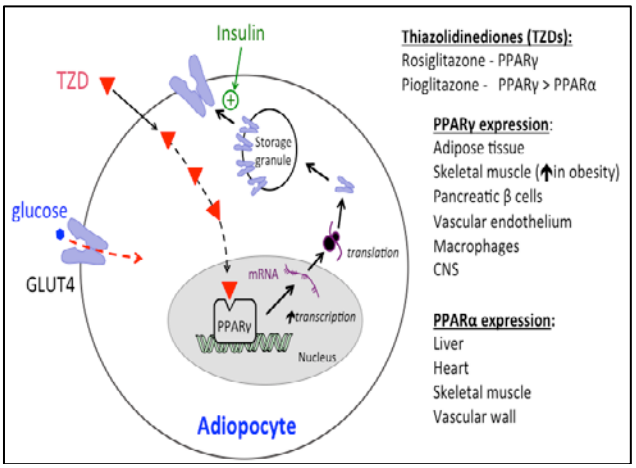

Figure 3: Glycemic mechanism of action of thiazolidinedione "insulin sensitizers"

MATERIALS AND METHODS

All the chemicals (reagents and solvents) were purchased from commercial suppliers (Merck grade) SigmaAldrich,Avra, and SD Fine Chem. Ltd and they were used further without purification.

Melting Point Apparatus

The melting points of the synthesized compounds were taken in the open capillary tubes using Chemline company CL726 melting apparatus.

Thin Layer Chromatography

Purity of the compounds was checked by TLC using silica gel G (0.5mm thickness) coated over glass plate (12 x 20 cm). For the determination R_f value the dried silica gel G coated over glass plate were used.

Preparation of TLC plate: By using distilled water silica gel G slurry is prepared and poured on to a glass plate which is maintained on a level surface. The slurry is spread uniformly on the surface of the glass plate. After setting, the plates are dried in an oven at 50ºC or 15 minutes for activating the TLC plate. Chromatogram was developed by ascending technique when solvent front travelled appropriate distance; plates were taken out and dried. The location of spot was detected using iodine chamber.

R_f = Distance travelled by solute / Distance travelled by solvent

Infrared Spectroscopy

The IR Spectra of the synthesized compounds were recorded at RBVRR women's College of Pharmacy by Shimadzu-FT/IR spectrophotometer in KBr disc. The IR value was measured in cm^{-1}.

Nuclear Magnetic Resonance

The H-NMR Spectra of the synthesized compounds were recorded at Central Facilities for Research and Development, Osmania University, Hyderabadby Bruker 300 MHz FT- NMR using $CDCl_3$(Deuteriated Chloroform) as internal standard. The PMR (Proton Magnetic Resonance) spectroscopic values are measured in δ ppm in DMSO-d6.

Mass Spectroscopy

Mass spectra was recorded in Schimadzu Mass Spectrometer.

Computational methods

Virtual screening studies:

To compare our *in silico* screen with a real *in vitro* experiment, we'll use Sulfonylurea Receptor (SUR-1) (PDB ID: 6jb3) as our target.

Exercise 1: Prepare the Input Structures

Click on File -> Import, click Next, enter 6jb3 for the PDB ID, and click Finish. Right-Click on 6jb3 in the Molecules tab, and select AutoDock -> Make Macromolecule. (1) create a 6jb3 folder under Macromolecules and (2) put 6jb3.pdbqt in it. Right-Click on any of the entries inside the Open Babel widget, and select Convert All to AutoDock Ligand (pdbqt). This opens a progress dialog box and shows the pdbqt files created in the Ligands folder. Now that we have the input pdbqt structures, we are ready to use the Vina Wizard.

Exercise 2:Using the Vina Wizard

Click on the Vina Wizard tab under the Controls pane. The first page for the Vina Wizard is similar to the AutoDockWizard.Click on the Start button to begin. On the Select Molecules page, use Shift + the mouse to select all the ligands, select 6jb3 under the Macromolecules folder, and click Forward. On the Run Vina page, click the Maximize button to make the Vina search space large enough to include all the atoms from our target. Click

Forward to start the virtual screen. PyRx now loops through all the selected ligands and runs Vina for each of them.

Exercise 3: Comparing Docking Results

PyRx stores virtual screening results in a multi-table database that you can access by clicking on the Tables tab under the View pane. This table widget provides a bird's-eye view of your workspace, and it includes tables for Ligands, Targets, and Docking Results.

Molecular docking studies:

1. Ligands preparation

Draw the structure of the ligand using chemsketch software, generate SMILES notation copy the SMILES notation of the compound and convert into .pdb format by using OPEN BABEL GUI software.In AUTODOCK vina software choose the **Input** option and select the ligand molecule from the folder.Choose**select Torsions** options and select the number of torsions in the ligand molecule. It should not be more than 8.Save the ligand molecule by using **Output** in pdbqtformat.A text document containing the name of the receptor, ligand and the position and dimensions of the grid box must be noted down.

2. Preparation of receptor

The crystal structure of the necessary protein is searched and downloaded from RCSB protein data bank in PDB format.Autodock app from Autodock tools is opened and the following steps are followed.Open the application.Choose**read molecule** option from **file** menu and open the downloaded protein from RCSBPDB.Add polar hydrogens to the protein from**Edit** menu and save it.Choose**macromolecules** from **grid** menu and choose it and it will converts as the macromolecule.Choose**grid box** option from **grid** menu and save the molecule in pdbqt format. Copy the file path of VINA folder and paste it in the command line. The command starts running and docking procedure is automated. The results are generated in the form of a text document and in a .pdbqt format file.

3. Docking of the ligands with the receptor using AutoDockVina

The .pdbqt file is used in order to analyze the results and identify the presence of hydrogen bonds in between the molecule and the receptor. Choose the **Analyze** option from the quick access tool bar and select **docking** option. Click on the **Open Autodock vina result** option and choose the ligand pdbqt output file. Then again click **Analyze> Macromolecule > open,** and select the protein molecule.Now again select the **Analyze**button, choose **docking** option and click **show interactions** option. Dialogue boxes which show the interaction between the molecule and the amino acid will appear on the screen along with the image.

RESULTS AND DISCUSSION

Virtual screeningResults of Potent HITS

Virtual screening of 100 molecules were performed by retrieving the imidazole molecules from CHEMBL database for various activities like anti-cancer, anti bacterial, anti fungal, anti diabetic.Among all the activities performed, imidazolones were found to have interesting anti diabetic activiteand these Imidazolones are the bioisosteres to the thiazolidinones and demonstrated both SUR-1 &PPARɣ agonistic activity. By assessing PPARɣ agonistic activity the molecules can be screened & molecules of highest scores were selected for further studies by using molecular autodock. The potent molecules having same antidiabetic activity those are shown in table 1.

From the results the Imidazole moieties as shown in Figure 4 with both SUR-1(PDB ID: 6JB3) &PPARɣ(PDB ID: 6fzy activity were identified as HITS.

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Table :1 Virtual screening results of SUR-1 & PPARɤ for Antidiabetic activity with Imidazoles

S.NO	CHEMBL ID	SCORES FOR (SUR-I)SULFONYL UREA RECEPTOR-1 TARGET(6JB3)	SCORES FOR (PPARɤ) PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR GAMMA TARGET(6FZY)
10	CHEMBL2385551	-10.1	-8.1
42	CHEMBL559966	-10.2	-8.2
46	CHEMBL118281	-10.1	-8.1
51	CHEMBL567258	-11.0	-8.5
65	CHEMBL261221	-10.5	-8.2
72	CHEMBL1778259	-10.3	-8.3
80	CHEMBL2234969	-10.3	-8.1
93	CHEMBL342119	-10.5	-8.4

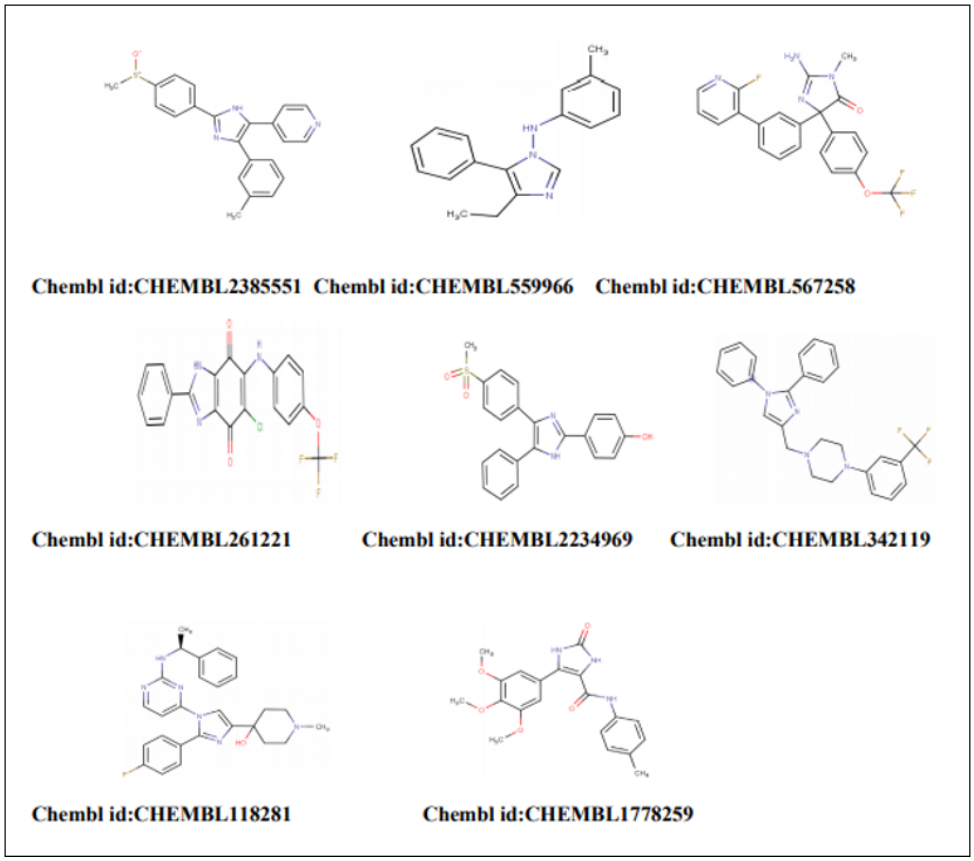
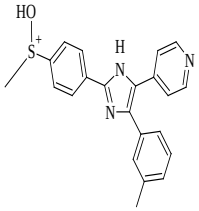
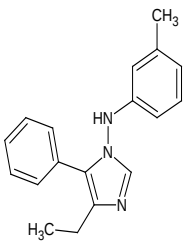


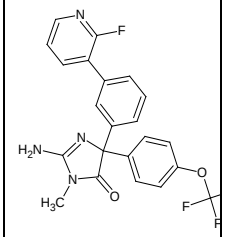
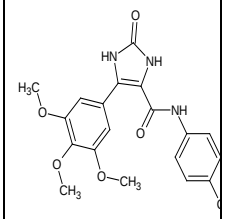
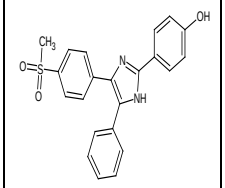
Figure 4: Structures of potent HITS choosen by virtual screening

Molecular Docking Results of Potent HITS

As the binding affinity studies between ligands and their receptors form the basis of physiological activity and pharmacological effects of chemical compounds. We carried out docking studies of five HITS obtained from virtual screening to investigate the correct binding pose of the newer molecules with SUR-1and PPAR-γ in order to assess their potency in antidiabetic activity.

Table 2: Molecular Docking interactions of 5 potent HIT molecules with SUR-1 (PDB ID: 6JB3) & PPARγ (PDB ID: 6fzy)obtained from virtual screening results

S.NO	STRUCTURE OF LEAD MOLECULES	MOL.INTERACTIONS				SCORES	
		SUR-1 (6JB3)		PPARγ (6fzy)		SUR-1 (6JB3)	PPARγ (6fzy)
		H-bonding	Other interactions	H- bonding	Other interactions		
1		None	**Hydrophobic interactions** ILE381,LEU1241,PHE433,TRP430,ILE1307,GLN1308,GLN1190, ALA1303. **Hydrophilic interactions** ASP1304.	None	**Hydrophobic interactions** VAL431,LEU405. **Hydrophilic interactions** LYS432,LYS466,GLU345.	-8.7	-7.0
2		None	**Hydrophobic interactions**ALA1303,GLN1190,GLN1191,ILE1307. **Hydrophilic interactions** SER1186,ARG1187,ARG1300,ARG1187.	None	**Hydrophobic interactions** ILE484,LEU481,GLN482,LEU493, LEU504. **Hydrophilic interactions** TYR501,ASP503.	-7.9	-8.0

S.NO	STRUCTURE OF LEAD MOLECULES	MOL.INTERACTIONS				SCORES	
		SUR-1 (6JB3)		PPARγ (6fzy)		SUR-1 (6JB3)	PPARγ (6fzy)
		H-bonding	Other interactions	H- bonding	Other interactions		
3		None	**Hydrophobic interactions**PHE591,PHE638,LEU481. **Hydrophilic interactions** THR540.	None	**Hydrophobic interactions** ILE484,VAL475. **Hydrophilic interactions** LYS485,GLN482,LYS 502.	-8.7	-7.4
4		None	**Hydrophobic interactions**MET429,PHE433,LEU124 1. **Hydrophilic interactions** GLN1308, SER1238, GLN1190.	None	**Hydrophobic interactions** LEU481,ILE484,LEU4 93, VAL478. **Hydrophilic interactions** GLN479,ASP503,GLN 482,TYR501	-7.5	-7.1
5		None	**Hydrophobic interactions** ILE1307. **Hydrophilic interactions** GLN1308.	None	**Hydrophobic interactions** PRO394. **Hydrophilic interactions** GLN479,GLU476,ASP 390.	-7.5	-7.4

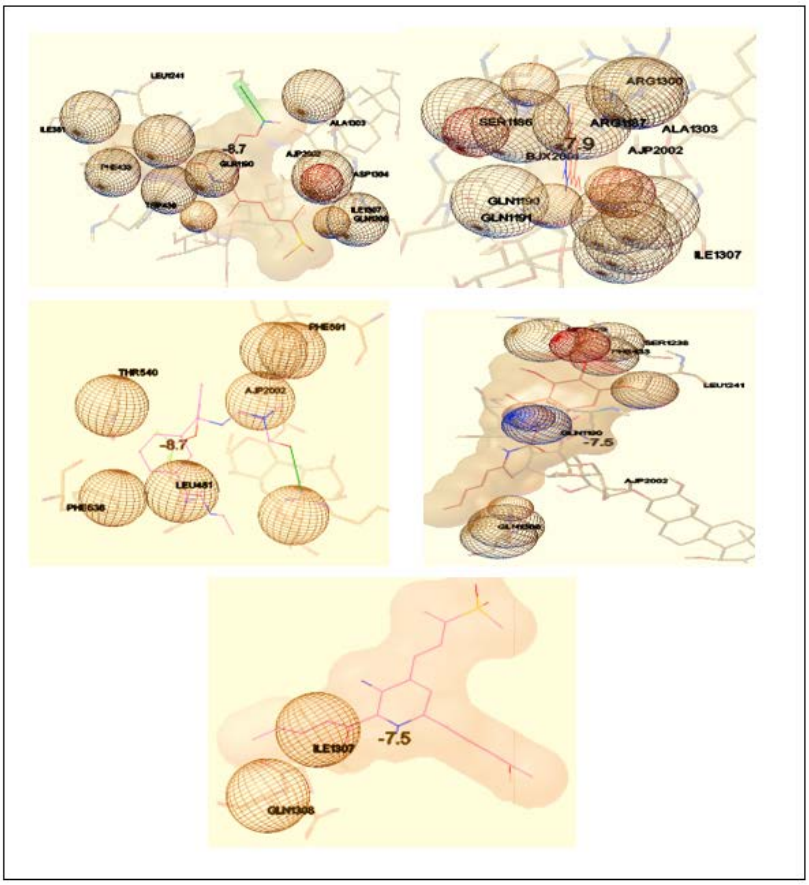


Figure5: Binding poses of 5 HIT molecules with SUR-1 (PDB ID: 6JB3)

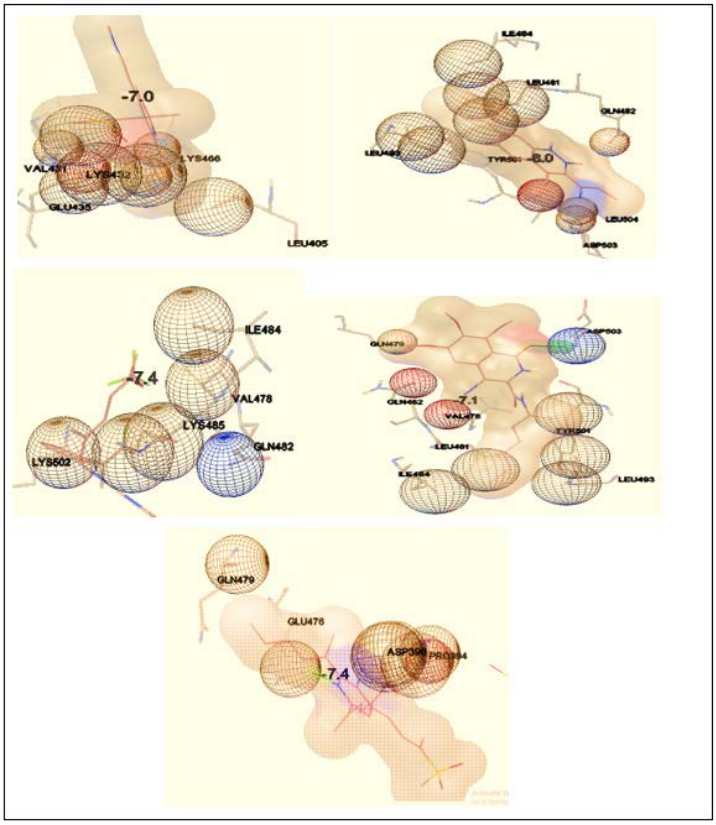


Figure 6: Binding poses of 5 HIT molecules with PPARγ (PDB ID: 6fzy)

DESIGN OF THE LEAD MOLECULE FROM SIMILAR STRUCTURESAND THEIR PHARMACOPHORIC FEATURES

By understanding the basic pharmacophoric features of the selected HITS, newer sulfonamido substituted imidazolones were designed.

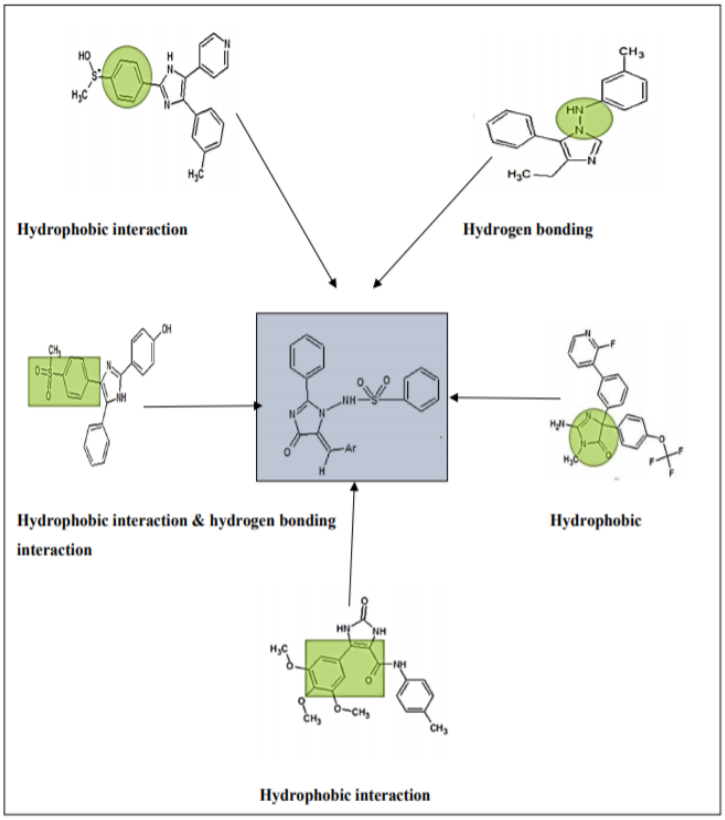
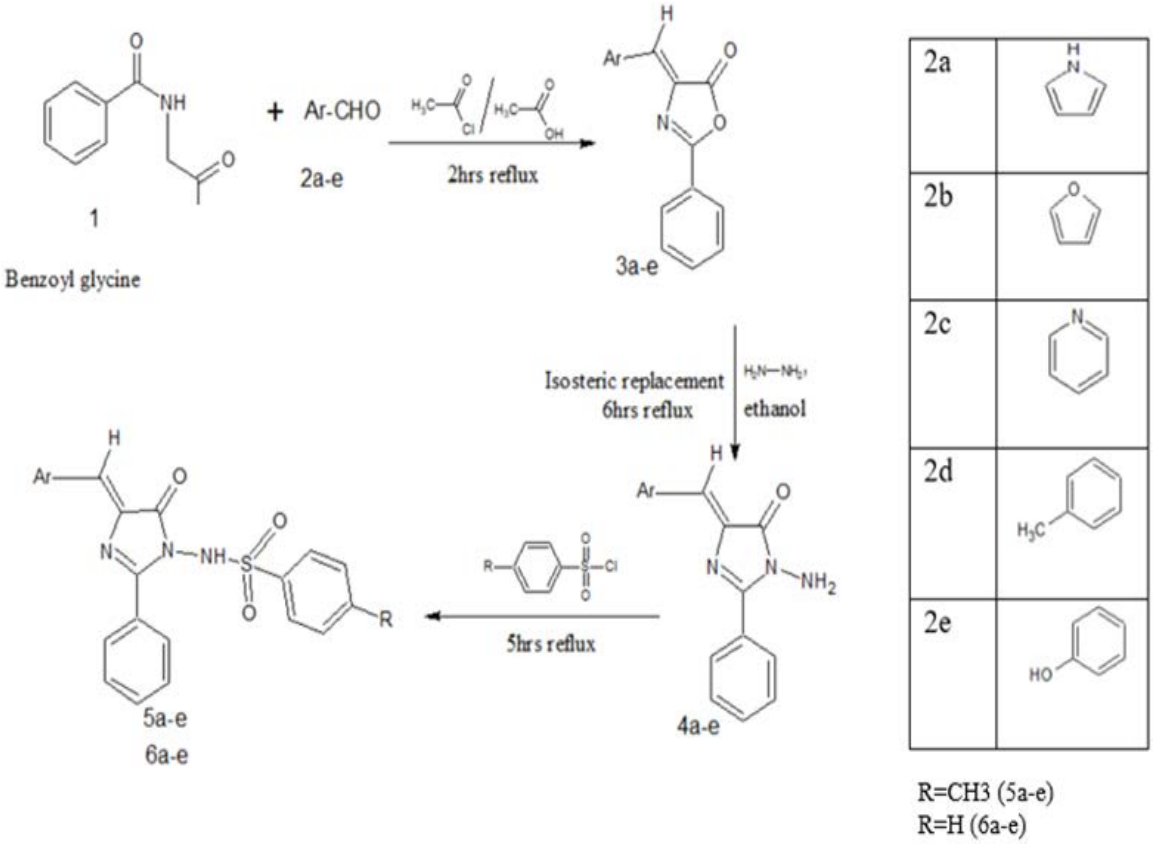


Figure 7: Design of the lead molecule

SCHEME

Synthesis of the designed molecules is achieved by the following scheme:



Molecular Properties and Toxicity Prediction of the Sulphonamido substituted imidazolone derivativesusing OSIRIS Property Explorer

On the basis of drug likeness, compounds were predicted to be promising druggable candidates.The toxicity of the compounds was also predicted using Osiris, most of the compounds amongst the synthesized ones showed non-tumorigenic and non-reproductive effects, which further supports the drug features in the molecules. This toxicity prediction would be useful for the selection of compounds to test in animal models.

lead optimisation using lipinski's rule:

1. A molecular weight less than 500.
2. Not more than 5 hydrogen bond donor groups.
3. Not more than 10 hydrogen bond acceptor groups.
4. A calculated log P value less than +5 (log P is a measure of a drug's hydrophobicity).
5. Molar refractivity from 40 to 130.

The rule describes molecular properties important for a drugs pharmacokinetics, including absorption, distribution, metabolism and excretion. It is to evaluate drug likeness or determine if a chemical compound with a certain pharmacological or biological activity has property that would make it likely orally active drug

Table 3: Molecular properties and toxicity prediction of the Sulphonamido substituted imidazolone derivativesusing OSIRIS Property Explorer&Molinspiration software

S.no	Log P	Log S	TPSA	natoms	Mol.wt	nON	nOHNH	NR	DL	MUT & REP
5a	2.82	-2.21	113.9	30	420.4	8	2	6	2.34	None
5b	2.92	-2.84	111.2	30	421.4	8	1	6	2.57	None
5c	2.77	-2.39	93.9	29	404.4	7	1	5	2.36	None
5d	4.39	-3.51	81.0	30	417.4	6	1	5	3.83	None
5e	3.44	-2.87	101.2	30	419.4	7	2	5	2.27	None
6a	3.27	-2.55	113.9	31	434.4	8	2	6	3.37	None
6b	3.37	-3.19	111.2	31	435.4	8	1	6	3.58	None
6c	3.22	-2.73	93.96	30	418.4	**7**	1	5	3.68	None
6e	3.89	-3.21	101.2	31	433.4	7	2	5	1.43	None

Pharmacokinetic Property Prediction of theSulphonamido substituted imidazolone derivatives

Pharmacokinetic properties of the title compounds were predicted using an online freeware SwissADME

Table 4: Pharmacokinetics Properties Prediction studies ofSulphonamido substituted imidazolone derivatives usingSwissADME software

Pharmacokinetic Properties	5a	5b	5c	5d	5e	6a	6b	6c	6e
GI absorption	High	High	High	High	High	High	High	High	High
BBB permeant	No	No	No	No	No	No	No	No	No
P-gp substrate	No	No	No	No	No	No	No	No	No
CYP1A2 inhibitor	No	No	No	No	No	No	No	No	No
CYP2C19 inhibitor	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
CYP2C9 Inhibitor	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
CYP2D6 Inhibitor	No	No	No	No	No	No	No	No	No
CYP3A4 inhibitor	No	No	No	No	No	No	Yes	No	No
Log K_p(skin permeation)	-6.67 cm/s	-6.48 cm/s	-6.64 cm/s	-5.73 cm/s	-6.25 cm/s	-6.50 cm/s	-6.31 cm/s	-6.47 cm/s	-6.08 cm/s

Molecular Docking of Sulphonamido substituted imidazolone derivatives with SUR-1 (PDB ID: 6JB3) &PPARγ (6fzy)

Molecular docking studies of the title compounds were carried out to understand the correct binding interactions of the title compounds with SUR-1 (PDB ID: 6JB3) &PPARγ (6fzy)

Table 5: Molecular Docking results of Sulphonamido substituted imidazolone derivatives using AutoDock Vina 1.5.6 software

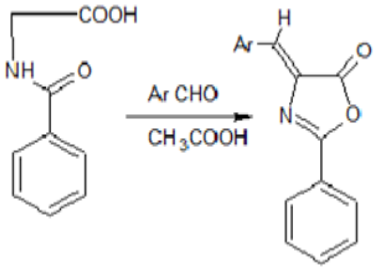
S.NO	IUPAC NAME OF LEAD MOLECULES	MOL.INTERACTIONS				SCORES	
		SUR-1 (6JB3)		PPARγ (6fzy)		SUR-1 (6JB3)	PPARγ (6fzy)
		H-bonding	Other interactions	H-bonding	Other interactions		
5a	*N*-{(4*Z*)-5-oxo-2-phenyl-4-[(1*H*-pyrrol-2-yl)methylidene]-4,5-dihydro-1*H*-imidazol-1-yl}benzenesulfonamide	ASN1245	**Hydrophobic** ILE1307,ALA1363. **Hydrophilc**GLN1308,ASP1304,ARG1167,GLN1191,GLN1190,SER1186,ASN1245,THR1242.	None	**Hydrophobic** LEU493,VAL478,LEU481.**Hydrophilic.**GLN498,LYS485.	-8.1	-7.4
5b	*N*-{(4*Z*)-4-[(furan-2-yl)methylidene]-5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1-yl}benzenesulfonamide	None	**Hydrophobic** ILE1307,PHE1182. **Hydrophilic**GLN1190,ASN1245,ARG1300,GLN1191,GLN1303,ARG1187.	None	**Hydrophobic** LEU504,LEU481.**Hydrophilic**GLN498,TYR501,LYS485,GLN482.	-9.9	-7.6
5c	*N*-{(4*Z*)-5-oxo-2-phenyl-4-[(pyridin-2-yl)methylidene]-4,5-dihydro-1*H*-imidazol-1-yl}benzenesulfonamide	ARG598	**Hydrophobic** LEU481,ILE544,PHE591.**Hydrophilic** ASN1301,ARG1145,SER1138,ASN547,ARG598.	GLN498	**Hydrophobic** VAL478,LEU493,LEU481.**Hydrophilic** GLN482,LYS502,GLN498,TYR501,LYS485.	-7.7	-7.5

S.NO	IUPAC NAME OF LEAD MOLECULES	MOL.INTERACTIONS				SCORES	
		SUR-1 (6JB3)		PPARγ (6fzy)		SUR-1 (6JB3)	PPARγ (6fzy)
		H-bonding	Other interactions	H-bonding	Other interactions		
5d	*N*-{(4*Z*)-4-[(4-methylphenyl) methylidene]-5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1-yl}benzenesulfonamide	None	**Hydrophobic** PHE1182,ILE1307,PHE433,TRP430. **Hydrophilic** THR1242,ASN1245,SER1188,ARG1300, GLN1190,ARG1187.	None	**Hydrophobic** VAL431,GLY427,PRO426. **Hydrophilic** GLU397,GLN472,LYS401.	-7.8	-7.7
5e	*N*-{(4*Z*)-4-[(3-hydroxyphenyl) methylidene]-5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1-yl}benzenesulfonamide	GLN369 ,ASN1296.	**Hydrophobic** LEU1289,GLY313,MET1290.**Hydrophilic** ARG370,GLN369,GLU1253,ASN1296,GLN444,ASN1293,HIS584,THR588,TYR377,ARG306.	None	**Hydrophobic** MET491,LEU493,LEU481. **Hydrophilic** LYS485,ASP503,LYS502, TYR501.	-8.2	-7.8
6a	4-methyl-*N*-{(4*Z*)-5-oxo-2-phenyl-4-[(1*H*-pyrrol-2-yl)methylidene] -4,5-dihydro-1*H*-imidazol-1-yl}benzene-1-sulfonamide	None	**Hydrophobic** ALA1303,PHE433,LEU1241.**Hydrophilic** ASN1245,GLN1190,ARG1300,ARG1187 ,ASP1304,GLN1308,ASN124.	None	**Hydrophobic** LEU481,MET491,PHE310,LEU493.**Hydrophilic** TYR501,SER492.	-7.6	-7.8
6b	*N*-{(4*Z*)-4-[(furan-2-yl)methylidene] -5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1-yl}-4-methylbenzene-1-sulfonamide	None	**Hydrophobic** LEU1248,LEU1241,PHE433,TRP430. **Hydrophilic** ASN1245,ARG1300,ARG1187,GLN119.	None	**Hydrophobic** LEU481. **Hydrophilic** GLN482,GLN498,TYR501,LYS502,ASP503.	-8.2	-7.9
6c	4-methyl-*N*-{(4*Z*)-5-oxo-2-phenyl-4-[(pyridin-2-yl)methylidene] -4,5-dihydro-1*H*-imidazol-1-yl}benzene-1-sulfonamide	None	**Hydrophobic** PHE536,LEU481,PHE591. **Hydrophilic** ARG598,SER543,ARG1145.	None	**Hydrophobic** LEU504. **Hydrophilic** GLN482,LYS485,TYR501,GLN498,ASP503.	-8.7	-7.4
6e	*N*-{(4*Z*)-4-[(3-hydroxyphenyl) methylidene]-5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1-yl}-4-methylbenzene-1-sulfonamide	SER1238.	**Hydrophobic** PHE433,LEU1241,MET429,ALA1303,TRP430,PHE1182. **Hydrophilic** SER1186,ASN1245,GLN1190,THR1242, ARG1300,SER1238.	None	**Hydrophobic** LEU493,MET491,LEU481.**Hydrophilic** LYS485,LYS502,TYR501.	-9.6	-7.8

EXPERIMENTAL

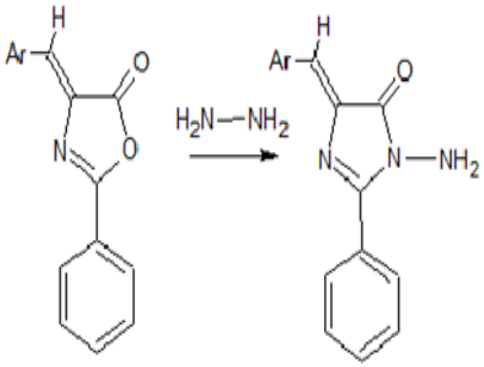
Experimental procedures

Step 1: General Procedure for the synthesis of (4Z)-4-(4-aryl benzylidene)-2-phenyl-1, 3-oxazol-5(4*H*)-ones.



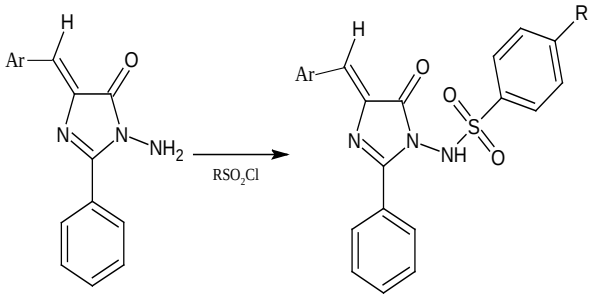
A mixture of aromatic aldehydes (2a-f)(0.25 moles), Hippuric acid(1a) (44.8 gm, 0.25 mole) / Benzoyl glycine (29 gm, 0.25 moles), anhydrous sodium acetate (15 gm), and acetic acid (59 ml) was heated at 110°C, with constant stirring. The mixture become almost solid, and then as the temperature rises, it gradually liquefies and turns deep yellow in colour. After completion of the reaction monitored by TLC the reaction is allowed to cool and ethanol (100 ml) is added slowly to the contents of the flask. After allowing the reaction mixture is left to stand overnight, the yellow colour product is filtered and washed with ice cold ethanol and finally with boiling water and recrystallized in ethanol. The crystalline products were then dried and characterized for various physio-chemical properties.All the derivatives were soluble in ethanol under hot condition. The yields were in a range of 70-75% with melting points ranging within 165-210˚C.

Step 2: General Procedure for the synthesis of (5*Z*)-3-amino-5-(4-arylbenzylidene)-2-phenyl-3,5-dihydro-4*H*-imidazol-4-one.



To a solution of compound (oxazolone) (0.0lmole) in 50 ml of absolute ethanol and hydrazine hydrate (0.03 mole), was added and the reaction mixture was refluxed for 6hrs. On cooling, the precipitate formed was filtered off, recrystallization by ethanol.The product obtained is very fine crystalline precipitate which is then dried on air to prevent the exhaustion of product under microwave oven and the yield obtained was 80-90% with the melting points ranging from 205-225˚C and the derivatives were freely soluble in water under normal room temperature.

Step 3:General procedure for the synthesis of*N*-[(4*Z*)-4-(arylmethylidene)-5-oxo-2-phenyl-4,5-dihydro-1*H*-imidazol-1yl]benzenesulfonamides.



Na_2CO_3 (2.785 g, 26.25 mmol) was added to a solution of imidazolone(12.5 mmol) in H2O (15 ml) at -5˚C to 10°C, followed by addition of p-Toulenesulphonyl chloride (2.86 g, 15 mmol) in three portions over a period of 1h. The slurry was warmed to room temperature and allowed to stir for 4h. Upon completion of the reaction which was monitored with TLC using CHCl3/CH3OH solvent system (9:1). The reaction mixture was acidified with 20% concentrated aqueous HCl solution to pH2, after which crystallization occurred and the product was obtained via suction filtration. And the yield obtained was 78-80% with the melting points ranging from 120-140 ˚C and the derivatives were freely soluble in water under normal room temperature. Compounds are freely soluble in ethanol.

Characterization of synthesized compounds:

(4Z)-4-(furan-2-ylmethylidene)-2-phenyl-1,3-oxazol-5(4H)-one 2b:

Black crystalline powder,yield – 63% , M.P: 160 – 200˚C characterized by the appearance of cyclic ester peak (C=O) (Lactone) stretching at1759cm^{-1}, C-H (s) peak at 2937cm^{-1},C=N(s) at 1674 cm^{-1}.

(5Z)-3-amino-5-(furan-2-ylmethylidene)-2-phenyl-3,5-dihydro-4H-imidazol-4-one 3b:

Light brown colourpowder, yield – 70% ,M.P: 130 – 140 ˚C characterised by the appearance of N-NH2(s) at 3059 cm^{-1},C=O (Lactone) at 1761 cm^{-1},C=N(s) at 1653 cm^{-1},C-N(s) at 1301 cm^{-1}.

N-{(4Z)-4-[(furan-2-yl)methylidene]-5-oxo-2-phenyl-4,5-dihydro-1H-imidazol-1-yl}benzenesulfonamide 5b:

Brown colourpowder,yield – 68% , M.P: 140 - 160 ˚C characterised by N-H(s) at 3342 cm^{-1}, C-H(s) at 2937 cm^{-1},C=N(s) at 1653 cm^{-1}, C-N(s) at 1180 cm^{-1}, S=O(s)-2PEAK at 1078,1415cm^{-1}, C=O (Lactone) at 1761 cm^{-1}.NMR values:^{1}H NMR: δ 1.25 (3H, s), 6.63 (1H, dd, *J* = 3.4, 1.8 Hz), 7.00 (1H, dd, *J* = 3.4, 0.9 Hz), 7.20 (1H, tdd, *J* = 7.2, 1.7, 1.4 Hz), 7.41-7.59 (4H, 7.54 (dddd, *J* = 7.8, 7.2, 1.5, 0.4 Hz), 7.54 (dddd, *J* = 8.0, 7.6, 1.5, 0.4 Hz)), 7.87 (1H, tt, *J* = 7.6, 1.5 Hz), 7.70-7.75 (3H, 7.75 (dd, *J* = 1.8, 0.9 Hz), 7.74(dtd, *J* = 8.0, 1.5, 0.4 Hz)), 7.97 (2H, dddd, *J* = 7.8, 1.5, 1.4, 0.4 Hz).

DISCUSSION

New series of (5,6a-e) were synthesized by a known convenient method. Sulphonamido substituted imidazolonederivatives were synthesized from3-amino-imidazol-4-one(5a-e). The compounds were characterized by IR, NMR and Mass Spectrometry.Sulphonamido substituted imidazolonederivatives were synthesized by isosteric replacement of various 4-aryl benzylidene-2-phenyl-1,3-oxazol-4-ones(3a-e)with hydrazine hydrate. 4-aryl benzylidene-2-phenyl-1, 3-oxazol-4-oneswere in turn prepared by the reaction of Benzoyl glycine with various aromatic aldehydes in the precents of anhydrous sodium acetate, and acetic acid. The synthesized compounds were then evaluated with physical and spectral data.

CONCLUSION

In the present investigation 100 molecules from CHEMBL database were retrieved and subjected to virtual screening against (PDB ID: 6JB3 & 6fzy). Five HITS with highest scores were selected and by understanding the basic pharmacophoric features, a new lead molecule was designed and from that a new series of Sulphonamido substituted imidazolones (5,6a-e) were subjected to molecular docking and toxicity prediction studies. All the title compounds were having potent anti-diabetic activity and were found to be safe. These molecules were synthesized and all the synthesized compounds characterized by physical and spectral data.

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