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Association of Antidiabetic Medication Targetting ppar-Gamma Agonists: Design, Synthesis & Evaluation

Ritu Sharma*, Mehul Zaveri, Gourav Jain, Neha Kawathekar
Department of Pharmacy, Shri G.S. Institute of Technology & Sciences, 23-Park road, Indore (M.P.) 452003, India. rtusrma@gmail.com,

Abstract: *Thiazolidinedione is an important target for Antidiabetic drugs. There is still significant potential for designing new chemical entity with affordable, safe and efficacious Antidiabetic drug. In present study 2-thioxo-4-thiazolidinedione derivatives were designed and interaction of these was investigated by docking studies in the binding site of PPARγ using (PDB ID 1FM9) enzyme using Glide v 5.6 (Schrodinger LLC., New York, USA; http://www.schrodinger.com). Among the series of designed compounds seven compounds with good potential were synthesized. Structural confirmation of these compounds was done by FT-IR, 1H-NMR and Mass spectroscopy. The compounds were evaluated for in vitro Antidiabetic activity against PPARγ agonists. The activity of compound RS4 and RS20 was found to be comparable with Pioglitazone. The above study could be very useful for further design and development of new Antidiabetic drug.*

Keyword*: Antidiabetic, Docking, PPARγ agonist, 2-Thioxo-4-thiazolidinedione*

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I. INTRODUCTION

Diabetes mellitus (DM) encompasses a group of metabolic diseases that has seriously threatened the human health and quality of life, due to its inherent complications, Alterations in carbohydrate, fat and protein metabolism are major contributing factors to DM, with consequential elevated blood glucose (hyperglycaemia), resulting from defective insulin metabolism. Recent reports have shown that DM is undoubtedly a rising global challenge, constituting major health risk in most countries, about 143 million people are estimated to be diabetic worldwide; and this number is projected to double by the year 2030 [1]. Precisely, type 2 diabetes mellitus (T2DM), is the most encountered form of DM associated with postprandial hyperglycaemia which accounts for about 80% of reported diabetic cases. A disproportion between reactive oxygen species (ROS) production and antioxidant scavenging capacity induces oxidative stress, which ultimately leads to cellular and tissue damage in diabetic individuals. These effects can be mitigated when oxidants are neutralized or scavenged by increased antioxidant supplementation [2]. The maintenance of moderate blood glucose level in Type 2 diabetic patients is largely achieved through the use of oral hypoglycaemic agents and insulin. However, these treatment options are expensive and have limited efficacy with significant adverse effects. Thus, continuous research with hypoglycaemic effect is imperative. Such researches will definitely offered and more efficacious therapeutic approaches for the treatment of diabetes and its inherent complications[3].

II. MATERIAL AND METHODS

2.1 Computational studies

2.1.1 Docking validation

The most eloquent method to check the accuracy of docking method is to determine the closeness between the lowest energy conformer and scoring function. Glide score simulate an experimental binding mode as deciphered by X-ray crystallography. Assurance of docking process was done by analyzing the RMSD value, it is used to indicate whether correct docking pose was obtained by Glide or not. Normally RMSD of 2Å and higher precision analysis is not meaning full [4]. Docking was done by using protein PPARγ using PDB ID 1FM9. The RMSD values among docked pose and its bound conformation for 1FM9 are in range of 0.002 to 0.048, which indicates that docking was performed well for PPARγ. After this validation, all of the twenty thioxo-zolidinedione inhibitors were docked in the binding pocket of X-ray crystallographic structure of 1FM9.

2.1.2 Ligand preparation

ChemDraw Ultra 12.0 was used to draw 2D molecular structures of designed hybrids. All these 2D structures were converted into 3D with help of Chem 3D ultra-version 8.0.3. These 3D structures were introduced into Maestro implemented in Schrödinger; energy minimization of 3D structures was done by using Ligprep v 2.4 programs. Different ionization states were generated at user defined pH. ConfGen was used to

generate various conformers for each ligand and minimization was done by using Impref module of Schrödinger suit by using OPLS-2005 force field to correct its bond length and bond order.

2.1.3 Protein preparation

The three-dimensional crystal structure of Protein (PDB ID 1FM9) was downloaded from RCSB Protein Data Bank and prepared by protein preparation wizard. Preparation and refinement are two components of protein preparation wizards. After confirmation of chemical accuracy, addition of hydrogens and side chain neutralization was done by using force field OPLS-2005. Only those side chains were neutralized that neither participate in salt bridge formation nor were present in contact with binding cavity. Minimization was performed until the average root mean square deviation of the non-hydrogen atoms reached 0.3 Å. In final step flip no flip model of prepared protein was obtained and this was used for grid generation.

2.1.4 Receptor Grid Generation

Receptor grid generation starts by picking and selecting co-crystallized ligand from the active site of prepared protein. Finally, the grid was generated to define the active site of protein which was visualized in form of box at the point of work space. The complete process was run by default settings. This grid file was further used to perform docking.

2.1.5 Docking studies

Ligand docking was done by using Glide, v 5.6. The prepared ligands and the file obtained from receptor grid generation panel were selected and all the designed hybrids of 4-aminoquinoline and acridine derivatives were docked within the binding site of PPARγ (PDB ID 1FM9). Flexible docking was done by employing Extra Precision (XP) mode of Glide. Glide score of compounds was obtained and various interaction of ligand with protein was studied. The final energy evaluation was done with the GlideScore and a single best pose was generated as output for a particular ligand with the help of following equation.

GScore = a*vdW+ b*Coul + Lipo + H bond + Metal + Bury P+ RotB + Site

Where vdW = Vander Waal energy, Coul = Coulomb energy, Lipo = Lipophilic contact term, HBond = Hydrogen-bonding term, Metal = Metal-binding term, BuryP = Penalty for buried polar group, RotB = Penalty for freezing rotable bonds, Site = Polar interaction at active site, and the coefficient of vdW and Coul are a = 0.065, b = 0.0130 [5]. The best pose for a given ligand was determined by the Emodel score, while different compounds were ranked using Glide score. The correlation of docking score with Glide Emodel energy was calculated and the value was found to be r^2=0.80.

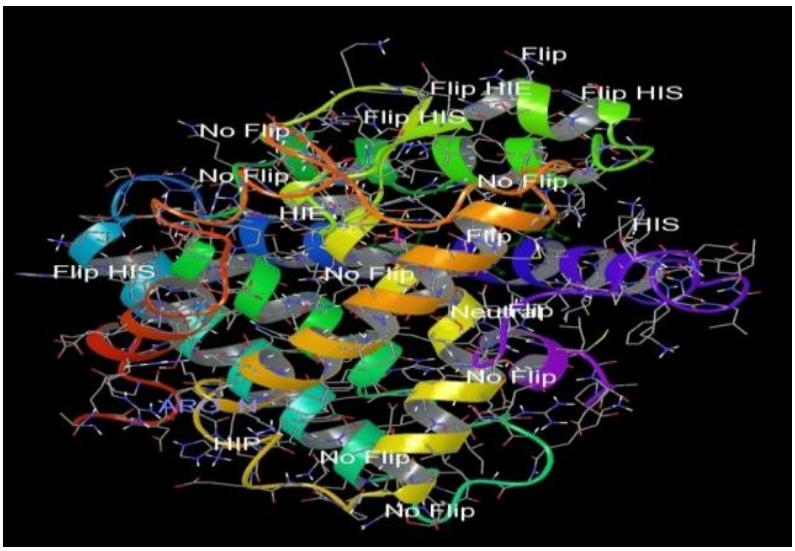


Fig.1 Image of Prepared Protein

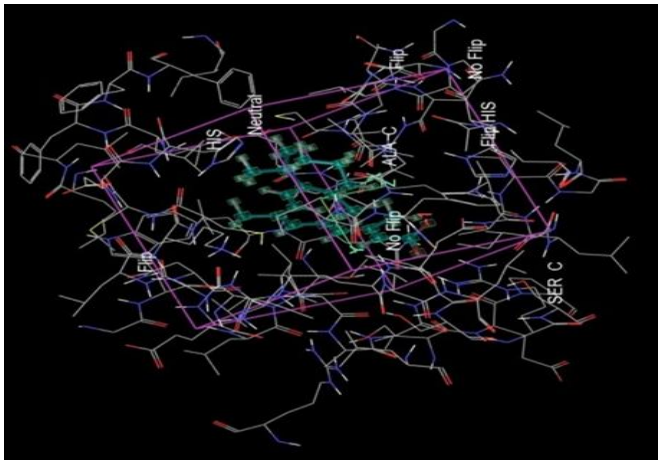
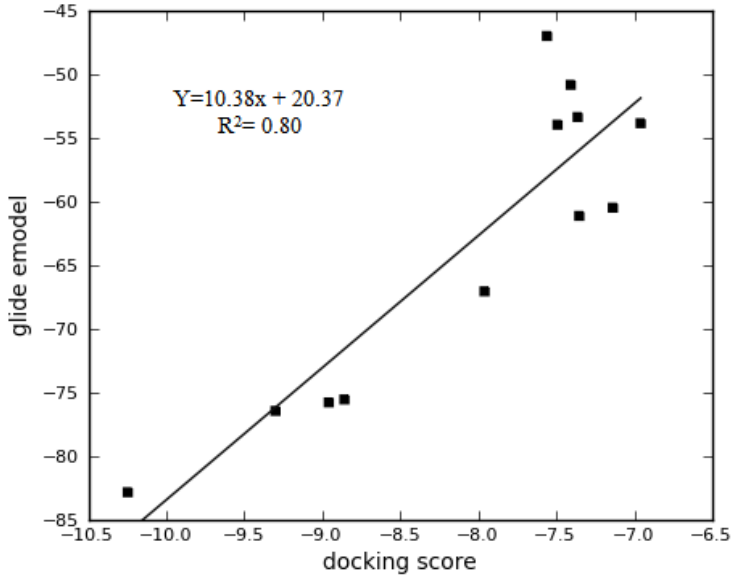
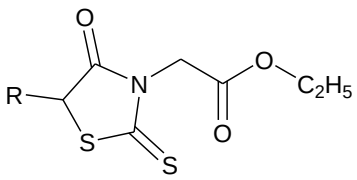


Fig.2 Image of Grid Generation



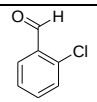
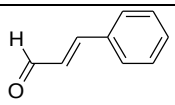
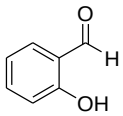
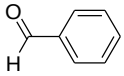
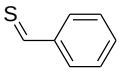
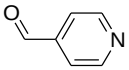
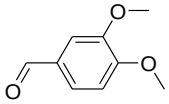
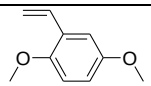
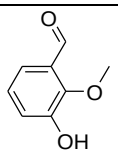
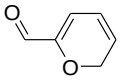
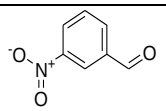
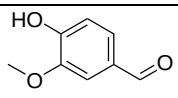
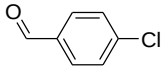
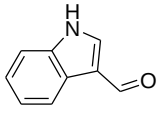
Graph 1: Correlation of docking score with glide emodel energy

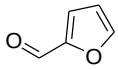
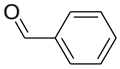
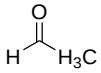
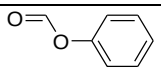
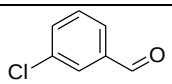
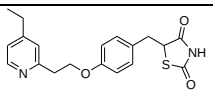


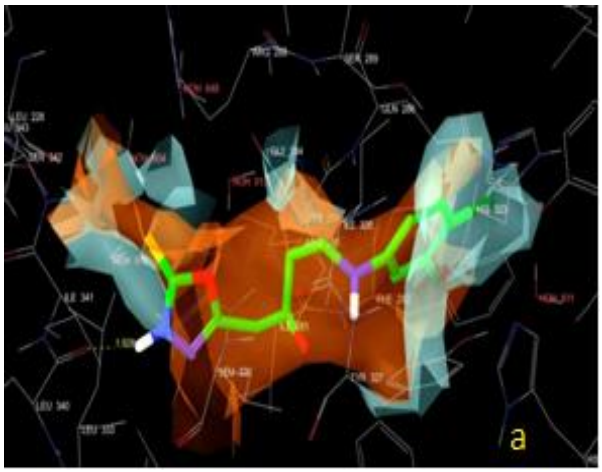
General Structure of Compounds RS1-RS7

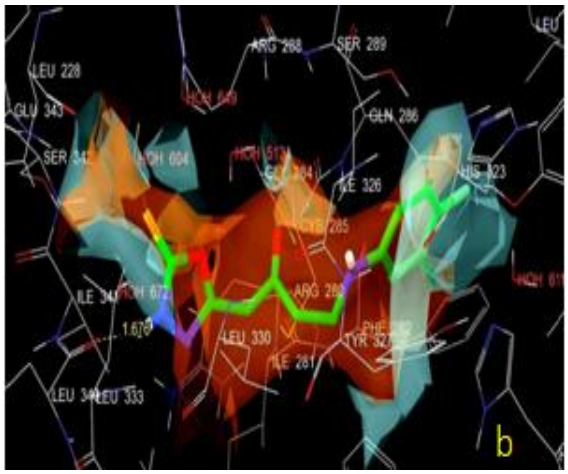
TABLE 1: Docking score and Glide emodel energy of compounds along with compound code, linkers (RS1-RS20)

Comp. Code for docking	R	Compound Name	Docking Score SP	Docking Score XP	Glide emodel

RS1		2 chloro Benz aldehyde	-7.2138	-8.9981	-42.1723
RS2		Cinnamaldehyde	-7.0091	-8.9871	-44.4813
RS3		Salicaldehyde	-7.0911	-8.9385	-47.5189
RS4		Benzaldehye	-7.0686	-8.7669	-57.8790
RS5		Thiopheneldehyde	-7.4715	-8.8151	-46.4260
RS6		Pyridine 4 carboxaldehyde	-7.3932	-8.0118	-43.6051
RS7		3-4 dimethoxyaldehyde	-7.6285	-8.7317	-28.3828
RS8		2-5 dimethoxyaldehyde	-7.2106	-8.3633	-39.2365
RS9		Hydroxy methoxy aldehyde	-7.2737	-8.7771	-45.4108
RS10		Paraldehyde	-8.1379	-8.8864	-48.2756
RS11		3 Nitro Benzaldehye	-7,8253	-8.4567	-364541
RS12		Vanillin	-7.8099	-8.0008	-48.4373
RS13		p-chloro Benzaldehye	-7.2102	-8.4321	-45.2341
RS14		Indole 3 carboxaldehyde	-7.2016	-8.3021	-42.1734

RS15		furfuraldehyde	-7.6693	-8.2669	-36.2892
RS16		Formaldehyde	-7.9178	-8.0528	-32.9323-
RS17		Tolualdehyde	-7.1291	-7.5964	-31.3558
RS18		Acetaldehyde	-7.2700	-8.5649	-39.8378
RS19		Anisaldehyde	-7.3711	-8.1298	-42.1324
RS20		3 chloro Benzaldehye	-7.1012	-9.1011	-43.7654
Pioglita-zone		Standard	-8.3740	-10.6027	-50.6772





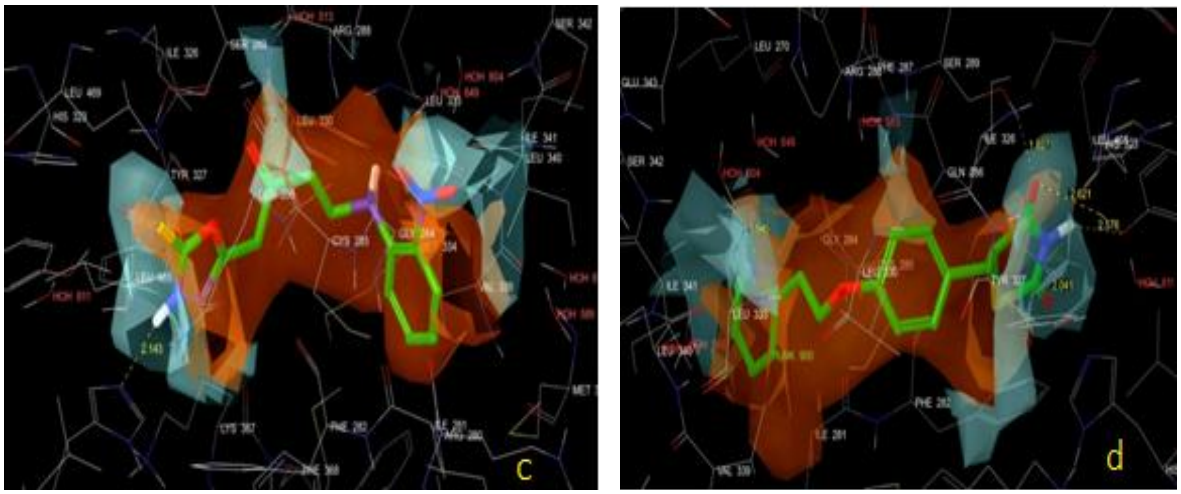


Fig 3: Extra precision Glide Docking with 1FM9 of compound (a) RS1 (b) RS4 (c) RS20 and (d) Pioglitazone.

TABLE 2: Docked conformation of designed compounds (RS1-RS20) with important amino acid residues and RMSD values

S no.	Compound code	Residues showing H-bond Interaction with 1FM9	RMSD
1	RS1	LEU 370, ILE 268,CYS 269	0.004
2	RS2	LEU 340, LEU 420, ILE 310	0.021
3	RS3	ILE 370, LEU 367, LUE 422	0.023
4	RS4	ILE 341, LEU 340, LEU 367	0.004
5	RS5	LEU 308, ILE 341, LEU 340	0.035
6	RS6	LEU 308, ILE 341, LEU 340	0.022
7	RS7	ILE 310, ILE 341, LEU 340	0.020
8	RS8	ASP 363, LEU 308, HIS 240	0.008
9	RS9	LEU 375, ARG 371, HIS 249	0.022
10	RS10	LEU 309, LEU 340, ILE 341	0.020
11	RS11	LEU 301, LEU 451,HIS 449	0.009
12	RS12	ILE 341, LEU 340, ILE 310	0.048
13	RS13	CYS 285,GLN 286, ILE 341,	0.002
14	RS14	ILE 341, LEU 340, LUE 276	0.031
15	RS15	ALA372, CYS 285,GLN 286	0.026
16	RS16	LEU308, GLN 286, SER312	0.002
17	RS17	ILE 341, LEU 340, LUE418	0.014
18	RS18	HIS449, CYS 369, LEU 309	0.024
19	RS19	SER 289, GLN 286,TYR 473	0.004
20	RS20	LEU 279, LYS 431, LEU 433	0.001

2.1.6 ADMET Prediction

Drug metabolism and pharmacokinetics play an important role in drug development. Absorption, distribution, metabolism and excretion are key parameters that are crucial for the success of a drug. Many drug candidates fail in the clinical trial due to poor ADMET profile. Thus it is important to carry out ADMET studies to minimize the chances of failure of the drug candidate in the clinical trials [6].

The ADMET studies were carried out in vitro using QikProp 3.3 module of Maestro v 9.0. Several descriptors that can be predicted by Qkiprop include Molecular weight, percentage human oral absorption, Log

P, Blood brain barrier penetration, Hydrogen bond donors and acceptors etc. The predicted ADMET properties of the synthesized compounds are reported in TABLE 3.

TABLE 3: ADMET prediction by QikProp

Comp. Code	Mole. weight	Qplog BB	QplogS	%Human Oral Absorption	Qplog Po/w	Rtvfg Reactive functioal group	Rule of five
RS1	329.00	-0.763	-3.430	90.748	2.247	0	0
RS2	321.10	-1.302	-3.766	88.109	2.470	0	0
RS3	311.10	-1.297	-3.364	80.583	1.596	0	0
RS4	295.10	-1.021	-3.061	86.135	1.795	0	0
RS5	295.00	-0.860	-2.982	85.715	1.716	0	0
RS6	296.10	-1.248	-2.042	75.466	0.735	0	0
RS7	355.10	-1.089	-3.335	89.329	2.048	0	0
RS8	355.10	-1.025	-3.117	89.692	2.014	0	0
RS9	341.10	-1.401	-3.089	78.056	1.443	0	0
RS10	325.10	-0.713	-3.328	85.713	1.753	0	0
RS11	340.30	-2.040	-2.627	83.704	0.864	0	0
RS12	341.10	-1.607	-3.390	75.424	1.448	0	0
RS13	329.00	-0.748	-3.496	90.215	2.277	0	0
RS14	331.10	-1.487	-4.347	80.491	2.167	0	0
RS15	285.30	-0.972	-2.050	81.568	1.032	0	0
RS16	219.30	-0.714	-0.934	78.010	0.172	0	0
RS17	321.40	-1.059	-3.676	88.074	2.123	0	0
RS18	233.30	-0.859	-1.567	78.203	0.446	0	0
RS19	325.10	-1.117	-3.224	86.501	1.861	0	0
RS20	329.00	-1.253	-3.744	91.332	2.379	0	0

The ranges of the predicted values are as follows:
1. Molecular weight (mol_MW) (219-355)
2. Octanol/water partition coefficient (Log Po/w) (0.8-3.8)
3. Aqueous solubility (QPlogS) (-1.5-5.4)
4. Brain/blood partition coefficient (QplogBB) (-0.71to -2.62)
5. Percent human oral absorption (75%-97%)

2.1.7 Site Mapping

Sitemap generates information on the character of binding sites using novel search and analysis facilities which provides information to maestro for the visualization of the sites.
The resultant van der walls and electric field grids are then used to generate the phobic and philic potentials. Using these potentials, sitemap partitions the accessible space in each site into the following three basic types of region:-

- Hydrophobic – region that are favorable for occupancy by hydrophobic ligand groups.
- Hydrophilic- region that is favorable for occupancy by hydrophilic ligand groups.

Neither hydrophobic nor hydrophilic- regions that are of mixed character or are far enough from the receptor surface to be similar to bulk water. Site mapping was performed and the results are shown in fig 4 .

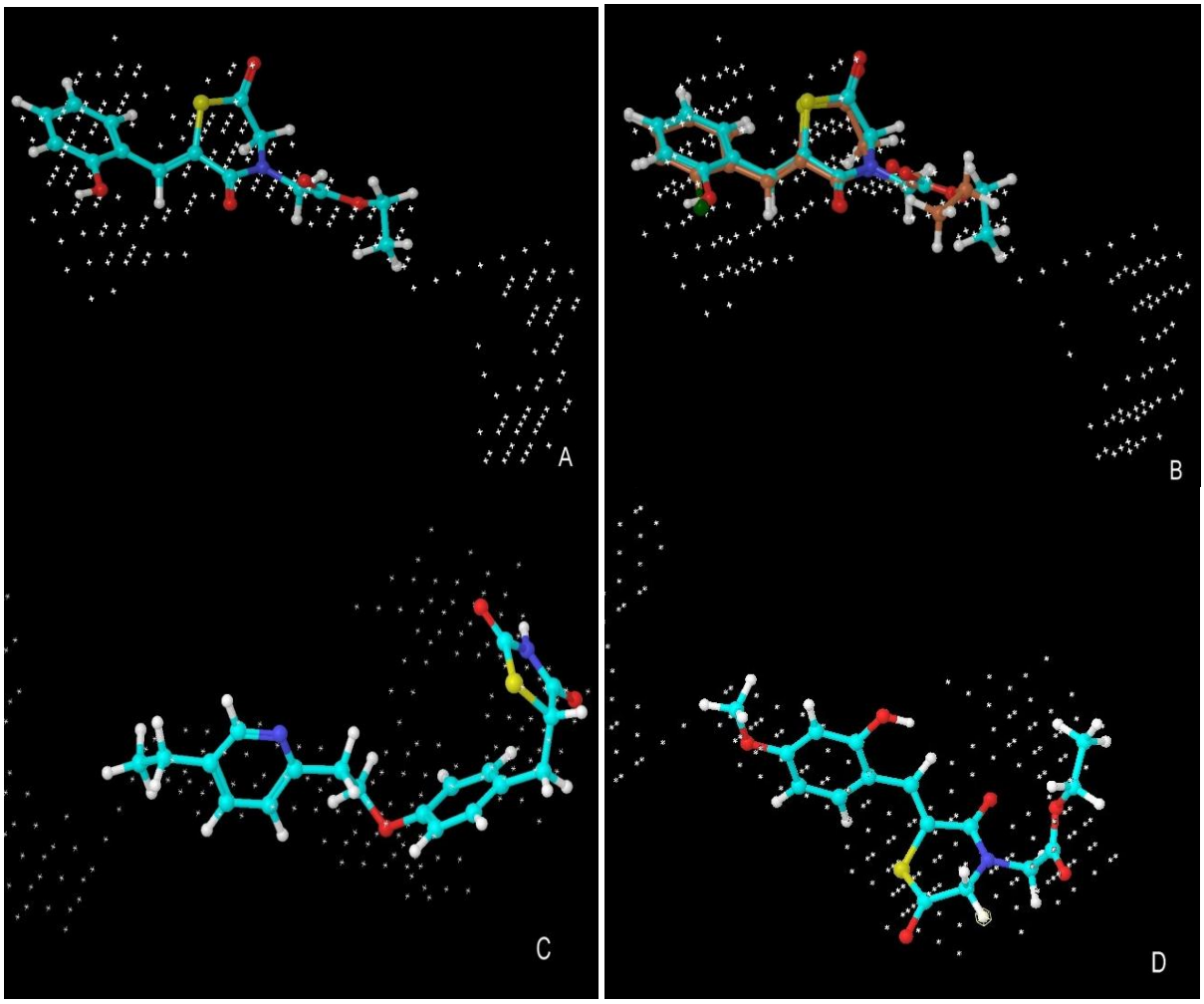
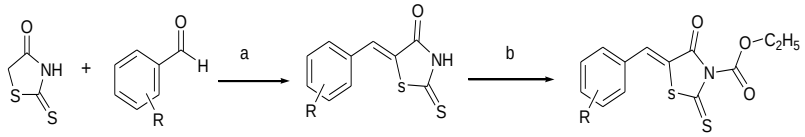


Fig 4: Hydrophobic, hydrophilic donor and acceptor maps of 1FM9 with co- crystallized ligand with inside pockets. a) RS1 (b) RS4 (c) RS20 and (d) Pioglitazone.

III. SYNTHESIS

Rhodanine and Derivatives of aldehyde were reacted in presence of Ethanol and Piperidine [7] to form 5-(substituted benzylidene)-5-thioxopyrrolidin-2-one (1). Later, 5-(substituted benzylidene)-5-thioxopyrrolidin-2-one was converted into 5-(substituted benzylidene)-2-oxo-5-thioxopyrrolidine-1-carboxylate-1-one (2) by reacting with Ethyl bromo acetate under basic conditions (NaH).



Rhodanine Derivative of Aldehyde Intermediate Product

Scheme 1. Reagents and conditions: (a) Piperidine, Ethanol, Acetic acid reflux for 24 hr. at 40-45˚C, (b) NaH, Ethyl bromoacetate, Dry DMF stir for 24 hr. at 40-45 ˚C.

3.1 General Procedure for Synthesis of Thioxazolidine-dione Derivatives.

3.1.1 Synthesis of 2-thioxo 4-thiazolidinedione derivative (RS1, RS2, RS20)

A Mixture of 2-thioxothiazolidin-4-one (2.4g.20 mmol), Benzaldehye derivative (2g. 20 mmol), Piperidine (1.4g 16 mmol) and Ethanol (150 ml) was refluxed for 16-24 hr. The reaction mixture was poured into H_2O and acidified with acetic acid (AcOH) to give derivatives of aldehyde -5-thioxopyrrolidin-2-one (intermediate) as solid, which were recrystallized from methanol [8]. Completion of reaction has been confirmed using TLC using Benzene: Ethyl acetate as solvent system (3:7)

Sodium hydride (0.576 g. 24 mmol) was added portion wise to a solutions of derivatives of aldehyde -5-thioxopyrrolidin-2-one (3g. 20mmol) in dry DMF and the mixture was stirred at 80^0c for 1.5 hr. The mixture was cooled to room temperature and a solution of ethyl bromoacetate (3.7 g 24 mmol) in dry DMF was added dropwise. After being stirred at 80^0c for 15-20 hr., the reaction mixture was poured into H_2O and the solid product filtered and recrystallized from EtOH/ H_2O Completion of reaction has been confirmed by TLC using Benzene: Ethyl acetate as solvent system (3:7)

3.1.2 Synthesis of 2-thioxo 4-thiazolidinedione derivative (RS4, RS7)

A Mixture of 2-thioxothiazolidin-4-one (1.8 g. 18 mmol), Benzaldehye derivative (2g. 20 mmol), Piperidine (1.8g 16 mmol) and Ethanol (100 ml) was refluxed for 16-24 hr. The reaction mixture was poured into H_2O and acidified with acetic acid (AcOH) to give Derivatives of Aldehyde-5-thioxopyrrolidin-2-one (intermediate) as solid, which were recrystallized from methanol. Completion of reaction has been confirmed using TLC using Chloroform: Benzene: Ethyl acetate as solvent system (2:4:4). Sodium hydride (0.48 g. 24 mmol) was added portion wise to a solutions of derivatives of aldehyde -5-thioxopyrrolidin-2-one (3g. 20mmol) in dry DMF and the mixture was stirred at 80^0c for 1.5 hr. The mixture was cooled to room temperature and a solution of ethyl bromoacetate (3.7 g 24 mmol) in dry DMF was added dropwise. After being stirred at 80^0c for 15-20 hr., the reaction mixture was poured into H_2O and the solid product filtered and recrystallized from EtOH/ H_2O Completion of reaction has been confirmed by TLC using Benzene: Ethyl acetate as solvent system (3:7)

3.1.3 Synthesis of 2-thioxo 4-thiazolidinedione derivative (RS5, RS10)

A Mixture of 2-thioxothiazolidin-4-one (2.4g. 20 mmol), Benzaldehye derivative (2g. 20 mmol), Piperidine (1.4g 16 mmol) and Ethanol (150 ml) was refluxed for 24-30 hr. The reaction mixture was poured into H_2O and acidified with acetic acid (AcOH) to give derivatives of aldehyde -5-thioxopyrrolidin-2-one (intermediate) as solid, which were recrystallized from methanol. Completion of reaction has been confirmed using TLC using Chloroform: Benzene: Ethyl acetate as solvent system (2:4:4). Sodium hydride (0.48 g. 24 mmol) was added portion wise to a solutions of derivatives of aldehyde -5-thioxopyrrolidin-2-one (3g. 20mmol) in dry DMF and the mixture was stirred at 80^0c for 1.5 hr. The mixture was cooled to room temperature and a solution of ethyl bromoacetate (3.7 g 24 mmol) in dry DMF was added dropwise. After being stirred at 80^0c for 20-26 hr., the reaction mixture was poured into H_2O and the solid product filtered and recrystallized from EtOH/ H_2O Completion of reaction has been confirmed by TLC using Benzene: Ethyl acetate as solvent system (3:7)

IV. RESULT AND DISCUSSION

4.1 SPECTRAL ANALYSIS

4.1.1 FTIR Spectroscopy

The Fourier transform Infrared spectroscopy of all the synthesized compounds were recorded on IR AFFINITY-1 1400 using KBr pellet technique were carried out in S.G.S.I.T.S, Indore and are expressed in cm^{-1}. The IR data of the all compound are summarized in TABLE 4

Table 4: IR data of the synthesized compound (RS1-RS20)

S.No	Comp. code	IR prediction cm^{-1}
1	RS1	1300 (C-O) sec. alcohol, 1820(C=O) carboxylic acid, 898(C=0)conj. ketone, 3268 (C-H)alkene,
2	RS2	3268(C-H) alkyne, 2928(C-H) alkene, 2853(C-H)Aldehyde, 2744 (C-H)aromatic compound, 1141(C-O)sec.alcohol, 897(C=C) alkene
3	RS4	3138(C-H) alkene, 2838 (C-H) alkene, 2821(C-H)aldehyde, 1085(C=O) Conjugated ketone, 1812(C=O) Carboxylic acid
4	RS5	3141(C-H) alkene, 2731(C-H) aldehyde, 2593(S-H) thiol, 1683(C=O) conj. Ketone, 1548(N-O) Nitro comp. 786(C-H) alcohol
5	RS7	3134(O-H)alcohol,3020(C-H) alkene,2756(C-H)aldehyde, 1883(C-H) Ar. Comp, 1808 (C=O)anhydride, 1684(C=O)ketone, 1312 (C-N) amine, 1250(C-N)amine,1207(C-O) ether, 679(C=C) alkene

6	RS10	2826(O-H) alcohol, 2674(C-H) carb. Group, 1880(C-O) Ar. comp. 1726(C=O) ester, 1617(C=C) ketone, 1432 (C-H) Alkene, 1217(C-O) ether, 953 (C=C) ketone
7	RS20	3353(C-H) alkyne, 2744(C-H) aldehyde, 2115(C-H) ar. Comp., 1814 (C=O) ketone, 1604(C=C) Unset. Ketone, 1227(C-N) amine, 1066 (C-O) alcohol,904 (C=C) alkene, 678(C-Cl) halogen

4.2 Mass Spectroscopy

Mass spectra analysis was recorded using a mass spectrometer with an ESI source as m/z fragmentation pattern for molecular ion peak determination. Mass spectroscopy was performed at IISER Bhopal by Agilent 7890A GC with 5975C MS system. Data of the all compound are summarized in TABLE 5.

Table 5: Mass spectral data of the compounds

S.No.	Code	Mol. Formula	Mol. Mass	Mass (m/z)
1	RS1	$C_{13}H_{12}ClNO_3S_2$	329	326
2	RS2	$C_{15}H_{15}NO_3S_2$	321	324
3	RS4	$C_{13}H_{13}NO_3S_2$	295	298
4	RS5	$C_{11}H_{11}NO_3S_3$	300	303
5	RS7	$C_{13}H_{13}NO_5S_2$	355	355
6	RS10	$C_{16}H_{19}NO_3S_2$	337	340
7	RS20	$C_{13}H_{12}ClNO_3S_2$	329	332

4.3 ^{1}H NMR Spectroscopy

^{1}H NMR spectra were recorded using VARIAN, USA Mercury plus 300 MHz NMR spectrometers using methanol as solvent at IIT Bombay and TMS as internal standard. Data of the all compounds are summarized in TABLE 6.

Table 6: ^{1}H NMR Spectroscopy

S.N.	Comp Code	Type of H	No. of Peak	Chemical shifts δ (ppm)
1	RS1	6	6	2.8 (CH_2Ar)3.1(CH_2Ar),4.9(CH_2X), 7.52(Ar-H),7.49(Ar-H),7.58(Ar-H),
2	RS2	5	5	8(Ar-H), 7.6(Ar-H),7.4 (AR-H),4.9(CH_2O),3.3(CH_2O)
3	RS4	4	4	5.1(C-H),4.7(CH_2O),3.6(CH_2O),3.2(CH_2X)
4	RS5	5	5	7.5(ArH),3.9(CH_2O),3.67($CH_2$2X),2.0(CH_2Ar),1.8(CH3)
5	RS7	5	5	7.55(Ar-H),7.1(Ar-H), 4.9(CH_2X), 3.9(CH_2O), 3.25(CH_2O)
6	RS10	6	6	8(Ar-H),7.9(Ar-H),4.8 (CH_2O),4(CH_2X),3.4(CH_2X),1.8(CH)
7	RS20	6	6	7.9(Ar-H),7.8(Ar-H),7.2(Ar-H),4.9(CH_2O),4.7(CH_2X),3(CH_2Ar)

V. DETERMINATION OF ANTI-DIABETIC ACTIVITY

Albino Wistar rats in each group were fasted overnight. The animals were divided into normolglycemic control, test groups, and reference group. Normal animals received saline solution (1mL/kg), test groups received a test solution (100 and 120 mg/kg), and reference group received pioglitazone (90mg/kg). Blood glucose determination was done at 0day (prior to any treatment) and 7day (after drug administration) it continued for 14 and 21 day [9]. Rats were housed in polypropylene cages lined with husk in standard environmental conditions (temperature 25±2◦C, relative humidity 55±10% and 12:12 light: dark cycle). The rats

were fed on a standard pellet diet ad libitum and had free access to water [9]. The experiments were performed after approval of the protocol by the Institutional Animal Ethics Committee (IAEC) and were carried out in accordance with the current guidelines for the care of laboratory animals.

5.1 Induction of type 1 diabetes (alloxan model)

Alloxan monohydrate was dissolved in saline and administered into fasted rats at a dose of 90 mg/kg (IP) body wt. The solution was fresh and prepared just prior to the administration. The rats were given 5% (w/v) glucose solution in feeding bottles for next 24 h in their cages to prevent hypoglycaemia after alloxan injection. After 72 h rats with blood glucose level greater than 200 mg/dl and less than 400 mg/dl were selected. The dose of the drugs induced to rats was 100 and 120 mg/kg. The treatment was continued for the next 4 days and blood samples were collected on 0^{th}, 7^{th}, 14^{th} and 21^{st} days after 1 h administration [10]. Blood glucose level (BGL) was estimated with the help of Dr. Morepen diagnostic glucose kit. Body weight of all animals was measured on the 0, 7^{th}, 14^{th} and 21^{st} days after 1 h of treatment. The biological evaluation of synthesized compounds is shown in TABLE 7. The percentage change of body weight was calculated from its initial weight.

$$\frac{\text{Initial Weight}}{\text{Final weight}} * 100$$

Table 7: Biological Activity of Thiazolidinedione derivatives

Code	Group	Blood Glucose Level(mg/dl)				% Red. In B.G.L
		0 day	7 day	14 Day	21 Day	
RS1	Control	345	340	320	315	91.30%
	Alloxen Induced	320	315	305	305	95.31%
	Test Solution 100mg/kg	330	300	260	200	60.60%
	Test Solution 120mg/kg	320	290	223	160	50.00%
	Standard (Pioglitazone)	340	290	230	148	43.52%
RS2	Control	340	332	316	302	88.82%
	Alloxen Induced	350	346	338	335	95.70%
	Test Solution 100mg/kg	367	345	295	120	32.69%
	Test Solution 120mg/kg	359	289	220	125	34.81%
	Standard (Pioglitazone)	315	280	232	136	43.17%
RS3	Control	289	282	279	275	95.15%
	Alloxen Induced	297	292	289	283	95.28%
	Test Solution 100mg/kg	315	280	203	136	43.17%
	Test Solution 120mg/kg	326	260	210	127	38.95%
	Standard (Pioglitazone)	336	269	201	129	38.39%
RS4	Control	345	340	320	315	91.30%
	Alloxen Induced	340	332	316	302	88.82%
	Test Solution 100mg/kg	320	290	223	160	50.00%
	Test Solution 120mg/kg	315	280	231	136	34.81%
	Standard (Pioglitazone)	367	296	180	120	32.69%
RS5	Control	289	283	271	275	95.15%
	Alloxen Induced	302	288	281	276	91.13%
	Test Solution 100mg/kg	315	274	223	134	42.53%
	Test Solution 120mg/kg	269	214	165	124	46.09%
	Standard (Pioglitazone)	324	285	206	129	39.81%
RS6	Control	295	287	275	270	91.52%
	Alloxen Induced	356	345	338	321	90.16%
	Test Solution 100mg/kg	312	245	178	129	41.34%
	Test Solution 120mg/kg	256	201	163	123	48.04%

	Standard (Pioglitazone)	265	196	142	113	44.14
RS7	Control	315	302	299	290	92.06%
	Alloxen Induced	302	289	280	276	91.13%
	Test Solution 100mg/kg	330	260	202	132	40.00%
	Test Solution 120mg/kg	310	283	180	126	40.64%
	Standard (Pioglitazone)	293	206	163	120	40.95%

VI. CONCLUSION

In the present study, we have reported docking, ADMET, synthesis and Antidiabetic activity of series of 2-thioxo- 4-thiazolidinedione derivatives. XP Glide docking scores and docking poses of designed compounds and standard suggest that these compounds adopt similar binding mode with active site residue of PPARγ as hydrogen bond, hydrophobic and π-π stacking interactions, which help in the stabilization of drug in active site. The *in vitro* evaluation of synthesized compound against pioglitazone.

The completion of reaction was monitored by TLC with solvent system as ethyl acetate: benzene (7:3). The structure elucidation was confirmed by IR spectroscopy, ^{1}H NMR technique and Mass spectrometry. The pharmacological evaluation of all the synthesized compounds was performed by using alloxan model of diabetes mellitus. Among the series of synthesized compounds RS4, 100 and 120 mg/kg dose gave 43.17% and 38.95% reduction in BGL respectively, they show the comparable result with standard (38.39%) same as RS20 100 and 120 mg/kg dose gave 40.00%, 40.14 % reduction in blood glucose level respectively, They are quite potent as they gave reduction in blood glucose level respectively. The standard drug being Pioglitazone. The compounds RS2, and RS7, has shown good reduction in blood glucose level whereas compounds RS1, RS5, and RS10 gave comparable results. Present study reveals that in near future, these compound could be developed as lead Antidiabetic analogs.

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