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Design, synthesis and biological evaluation of some substituted sulphonyl urea/ guanidine derivatives as hypoglycemic agents

Panchal Ishan*, Panigrahi Bibhuranjan, Modh Kamal and Patel C N

Shri Sarvajanik Pharmacy College, Mehsana, Gujarat, India

ABSTRACT

Design and synthesis of sulphonyl urea and guanidine derivatives as hypoglycemic agents based on the indirect molecular modeling approach by checking the superimposibility of the design molecule and reported compound n-(6-substituted-1,3- benzothiazol-2-yl)benzene sulfonamides. Synthesize compounds was characterize by TLC, UV spectra, IR spectra, Mass spectra & ^{1}H NMR. All the synthesized compounds were subjected to OGTT to gain preliminary information regarding the anti hyperglycemic effect in normal Albino wistar rats which are chosen for the study. Glibenclamide was chosen as the standard. Comparisiton of data was performed by one wary ANOVA followed by post test, Dunnett's multiple comparisiton tests.

Key words: Hypoglycemic agents, molecular modeling, OGTT, ANOVA.

INTRODUCTION

Diabetes is a chronic disease that occurs when the pancreas does not produce enough insulin or alternatively, when the body cannot effectively use the insulin it produces. Insulin is a hormone that regulates blood sugar. Hyperglycemia, or raised blood sugar, is a common effect of uncontrolled diabetes and over time leads to serious damage to many of the body's systems, especially the nerves and blood vessels.

Type 1diabetes previously known as insulin-dependent or childhood-onset is characterized by a lack of insulin production. Without daily administration of insulin, Type 1 diabetes is rapidly fatal.

Type 2 diabetes formerly called non-insulin-dependent or adult-onset results from the body's ineffective use of insulin. Type 2 diabetes comprises 90% of people with diabetes around the world, and is largely the result of excess body weight and physical inactivity.

Prevalence and Incidence

More than 180 million adults were living with diabetes globally [1]. In 2003, the total was 194 million.[2] By 2030, the figure is expected to rise to 366 million.[3]Type 2 diabetes accounts for approximately 90 percent of all diabetes cases.[1]Two people develop diabetes every ten seconds.[4]

Mortality and Complications

Every 10 seconds a person dies from diabetes-related causes [4] Diabetes is the fourth leading cause of global death by disease[4].Each year diabetes accounts for 3.8 million deaths [4]. An even greater number die from cardiovascular disease made worse by diabetes-related lipid disorders and hypertension [4]. Diabetes is responsible for approximately six percent of total global mortality, about the same as HIV/AIDS. Over time, diabetes can damage the heart, blood vessels, eyes, kidneys, and nerves.

Risk factors

In some people pancreas fail to produce insulin, which results in Type 1 diabetes. Most people fall prey to the disease because their body stops processing the Insulin produced by the pancreas. These people have type 2diabetes, also called Diabetes mellitus. Diabetes acquired during pregnancy, known as gestational diabetes, can develop because extra stress during pregnancy can cause high glucose levels. A small number of people develop diabetes when their pancreas gets destroyed due to an accident or injury. Family history, Obesity, Lake of exercises, Luxurious life style, Fast food, Smoking and alcohol drinking, Mental Stress.

Warning Signs of diabetes

Extreme thirst, frequent urination, Constant hunger, Blurred vision, Sudden weight loss, Nausea & vomiting, Infections & extreme tiredness, Feeling tired and lethargic, Slow-healing wounds, Itching and skin infections, Mood swing

Aim of work

Diabetes mellitus is a chronic metabolic disorder with multiple etiologies. Among them non insulin dependent diabetes mellitus is very common world wide. This disease is associated with reduced life expectancy significant morbidity due to specific diabetes related micro and macro vascular complications there by diminish the quality of life.

So there is growing need of effective therapies to achieve optimal glycemic control in management of diabetes. Number of orally administrated antihyperglycemic agents has increased significantly in last decade. However current therapies to reduce plasma glucose level have inherent problems including compliance, ineffectiveness and occurrences of hypoglycemic episode. Therefore there is a need for more effective orally administrated agents that will both normalize glucose and insulin level.

Hermenegilda Moreno-Dı´az et. al. has reported N-(6-substituted-1,3- benzothiazol-2-yl)benzene sulfonamides having antidiabetic potential [8].Based on the indirect molecular modeling approach by checking the superimposibility of the design molecule and reported compound (RMSD=0.124) we have planned to synthesize some sulphonyl urea and guanidine moiety and check their activity as anti hypoglycemic agents.

Table 1 various approaches for treatment of diabetes

Insulin or insulin mimetics[5]	
Insulin/modified insulin	Provides better glycemic control
Improved delivery vehicle	Shows more favorable pharmacokinetics
Insulin mimetic	Selectively activates the human insulin receptor
Enhancers of insulin release[6,7]	
Sulfonyl ureas	Act only in the presence of elevated glucose level
Glucagon like peptide	Stimulates b-cell growth and differentiation
Imidazoline	Potent effect on glucose tolerance
Inhibitors of hepatic glucose production	
Glucagon receptor antagonists	Non-competitive action with glucagon receptor
Glycogen phosphorylase inhibitor	Decreases glucose-1-phosphate formation from glycogen
Pyruvate dehydrogenase kinase inhibitor	Increases oxidative glucose metabolism and decreases gluconeogenesis
Fructose-1,6-biphosphatase inhibitor	Decreases Pyruvate conversion to glucose
Glucose-6-phosphatase inhibitor	Affects final step in gluconeogenesis
Inhibitors of glucose uptake	
Glycosidase inhibitor	Inhibits *a*-glycosidase and decreases conversion of fructose to glucose
Inhibition of gastric emptying	Moderate postprandial glucose spikes
Inhibition of Na+ glucose co-transporter (SGLT)	Blocks renal glucose reabsorption from urine, used to induce glycosuria
Enhancer of insulin action	
PPAR*a* agonist	Decreases obesity
PPAR*g* agonist	Lipid and cholesterol homeostasis
Retinoid X receptor	Controls lipid and carbohydrate metabolism
*b*3 Adrenergic receptor agonist	Decreases food consumption and leptin
Protein tyrosine phosphatase-1B inhibitor	Prevents dephosphorylation of activate insulin receptor
Glycogen synthase kinase-3 inhibitor	Activates glycogen synthase

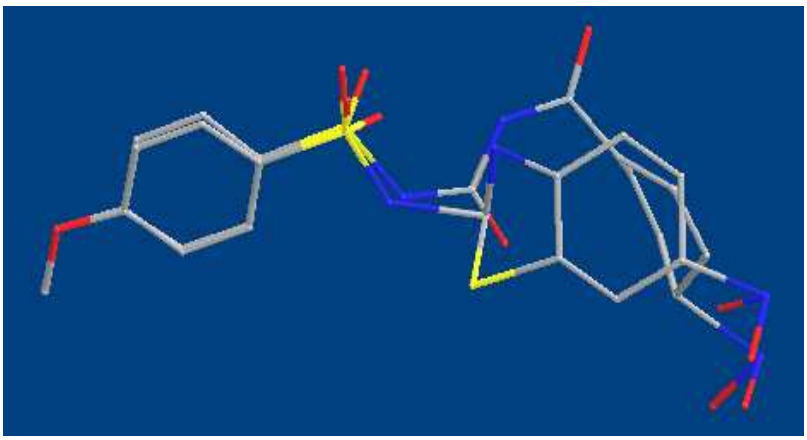


Figure 1 : Overlay of both structure in 3-D

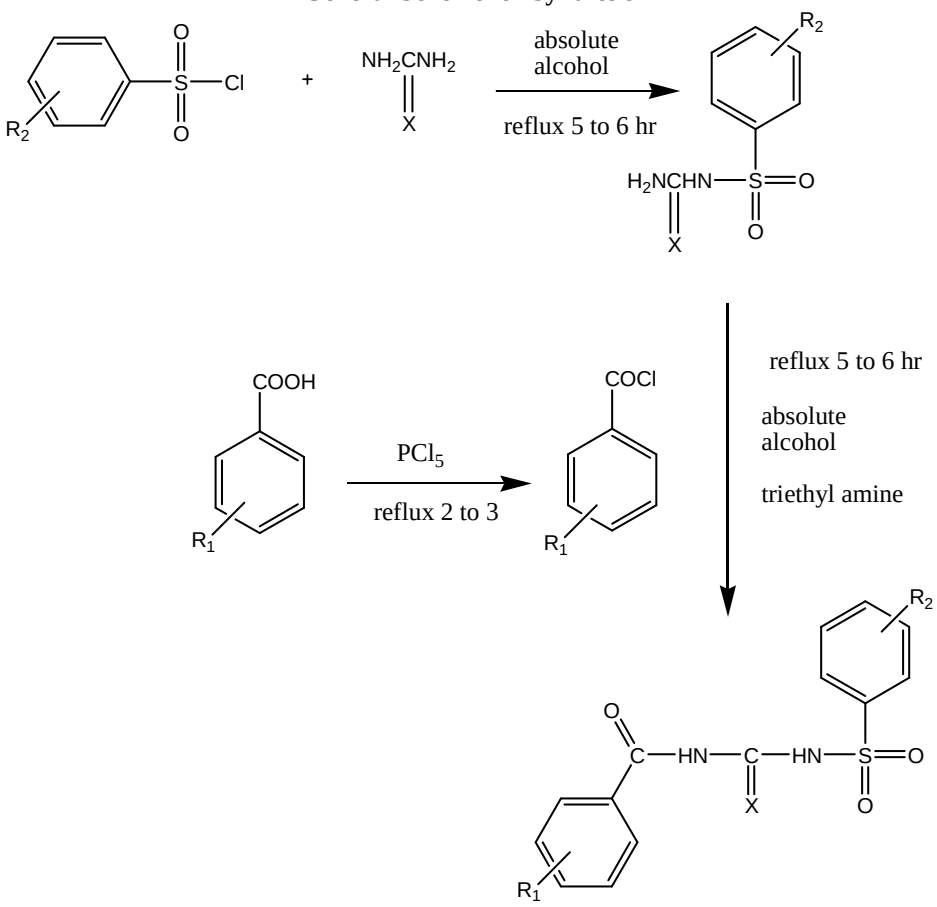
Chemicals

All the chemicals used for synthesis of title compounds were procured from S.D. fine chemicals Mumbai, Finar chemical Ltd. Ahmadabad and Loba chemical pvt. Ltd. Mumbai. The chemicals were used without further purification.

Instruments

- All the melting points were determined in open capillaries and are uncorrected.
- Thin layer chromatography was performed on microscopic slides (2×7.5cms) coated with Silica-Gel-G and spots were visualized by exposure to iodine vapor.
- UV spectra were recorded in absolute alcohol on UV-VIS 160A Shimadzu spectrophotometer.
- IR spectra of all compounds were recorded in KBr on FT-IR 8400S Shimadzu spectrophotometer using KBr.
- Mass spectra were obtained using 2010EV LCMS Shimadzu instrument.
- ^{1}H NMR spectra were obtained in $CDCl_3$ on BRUKER Advance-II 400 MHZ instrument and chemical shift were measured as parts per million downfield from Tetramethylsilane (TMS) as internal standard, chemicals shifts were expressed as δ value.

General Scheme for Synthesis



(1)

R_1 = - Cl, -NO_2, R_2= -H, -CH_3, X = O, NH

_____

***In vivo* study of synthesize compounds by OGTT** [9]

The oral glucose tolerance test (OGTT) measures the body's ability to use a type of sugar, called glucose that is the body's main source of energy. OGTT, a test of immense value and sentiment, in favor of using fasting plasma glucose concentration alone was seen as a practical attempt to simplify and facilitate the diagnosis of diabetes. Hyperglycemia is an important factor in the development and progression of the complications of diabetes mellitus.

Figure: 2 Blood collection from the retro orbital plexus

MATERIALS AND METHODS

A total number of 18 albino wistar rats weighing about 250-300 gm age were procured from animal house of Torrent research centre Gandhinagar. Group I served as a normal control group while group II for Glibenclamide control group. Group III- X were treated with synthesize compounds. The reference drug and the synthesize compounds were administered orally with oral feeding tube to the rats. OGTT for non diabetic rats were performed according to the standard method. In short, Group I to Group X was selected for OGT test after starving at water for 16 hours. The baseline glucose level was measured by Autobio-analyger having modeled number Microlab 300 (merck).

Group I stands for normal control group. Group II is treated with Glibenclamide (20 mg/kg body weight). The synthesize compounds was dissolve in DMSO (dimethlysulfoxide) according to 20 mg/ kg of body weight. Then the solution was administered orally to the glucose fed (2 gram/kg body weight) rats at the dose of 20 mg/kg body weight. Blood was collected from the retro orbital plexus. Then centrifugation of blood sample was done in research centrifuge with 4000 rpm speed having model number TC 4100D (eltek). Then the serum was collected with micropipette. Serum glucose was measured with Accucare glucose kit using GOD-POD method which was estimated by using Autobio-analyger at 0, 15, 45, 90, 120 minutes. Data were expressed as mean ± standard error of mean (SEM). Statistical comparisons were performed by one-way ANOVA followed by Dunnett's Multiple Comparison Test and the values were considered statistically significant when $P<0.05$.

Physical characteristics

All the synthesized compounds were off white to pale yellow and green colored crystalline solids. All the compounds are freely soluble in alcohol and other solvents like dichloromethane, methanol and acetone. The melting points of the compounds were in the range of 74 ºC to 224 ºC.

Spectral characteristics

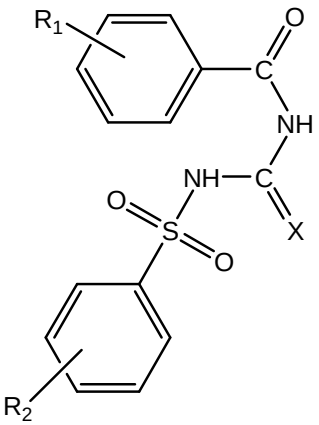
UV spectra: UV spectra were recorded in UV-1700 Shimadzu spectrophotometer. UV spectra of all the compounds were studied in ethanol. All the Compounds were found to have absorption λ_{max} in the range 261 nm to 381 nm.

IR spectra: IR spectra of all compounds were recorded on FT-IR 8400S Shimadzu spectrophotometer using KBr. All the synthesized compounds have shown characteristic carbonyl stretching at about 1680 cm^{-1} due to presence of amide group. All compounds have shown -S=O stretching vibrations at around 1130-1180 cm^{-1} (antisymmetric) and 1080-1200 cm-1 (symmetric).

1H NMR spectra: The ^{1}H NMR spectra of some of the compounds were studied in $CDCl_3$. All the compounds show characteristic chemical shift from TMS in terms of δ ppm. δ value obtained in the range of 7.2 to 8.2 signifies the presence of aromatic ring along with the δ at about 1.8-2.1 corresponds to the presence of methyl group.

Mass spectra: Mass spectra were obtained using 2010EV LCMS Shimadzu instrument. All the spectra were taken in positive and negative mode. The compounds show characteristic M+ and M+ 2 peaks.

General structure of compound synthesized



R_1 = NO_2, Cl, R2 = CH_3, H, X = O, NH

(1)

Table 2: Physical properties

Code	R_1	R_2	X	Molecular formula	Molecular weight (g/mol)	Melting point (˚C)	Yield (%w/w)	R_f value	Mobile phase toluene: methanol
1A	NO_2	CH_3	O	$C_{15}H_{13}N_3O_6S$	363.35	176-178	51.10	0.75	2:1
1B	Cl	CH_3	O	$C_{15}H_{13}ClN_2O_4S$	352.79	216-218	33.16	0.70	2:1
1C	NO_2	H	O	$C_{14}H_{11}N_3O_6S$	349.32	74-76	40.32	0.45	2:1
1D	Cl	H	O	$C_{14}H_{11}ClN_2O_4S$	338.77	88-90	34.48	0.65	3:1
1E	NO_2	CH_3	NH	$C_{14}H_{14}N_4O_5S$	362.66	68-70	42.73	0.78	3:1
1F	Cl	CH_3	NH	$C_{15}H_{14}ClN_3O_3S$	351.81	182-184	41.52	0.62	3: 1
1G	NO_2	H	NH	$C_{14}H_{12}N_4O_5S$	348.33	178-180	35.46	0.52	3: 2
1H	Cl	H	NH	$C_{14}H_{12}ClN_3O_3S$	337.78	188-190	43.17	0.48	2:1

Table 3: Spectral characteristics

Comp code	IR (cm^{-1})	Mass (m/e)	^1H NMR (δ ppm)	UV λ_{max} (nm)
1A	1530(NO_2) 1324(NO_2) 1685(CO) 1714(CO) 1374(CH_3)	365.1(M+2)	δ7.5-7.7(m,4H,Ar H) δ8.28(d,2H, NO_2ArH) δ8.21 (d,2H COArH) δ1.39 (s,3H,ArCH_3) δ4.5 (s,2H,NH)	295
1B	1680(CO) 1719(CO) 3139(NH) 815(Cl) 1369(CH_3)	353.6(M+1)	δ 7.1-7.7 (m,8H,Ar-H) δ1.25 (s,3H Ar-CH_3) δ 4.8 (s,2H,NH)	264
1C	1680(C=O) 3100(NH) 1530,1320(NO_2)	349.1(M+)	δ 7.5-7.9 (m,4H,Ar-H) δ 8.35(d, 2HCOArH)	315
1D	1690(CO) 3210(NH) 1705(CO)	338 (M+) 340 (M+2)	------	264
1E	1710(C=O) 3336(NH) 1533(NO_2) 1369(CH_3)	364(M+2)	δ7.3-7.8(m,4H,Ar H) δ2.25 (s,3H Ar-CH_3) δ8.35(d,2H, NO_2ArH)	308
1F	1677(C=O) 815(Cl) 3193(NH)	351(M+) 353 (M+2)	------	264
1H	1762(C=O) 1307(NO_2) 1523(NO_2)	333.1(M)	-------	264

In vivo antidiabetic activity by OGTT

Table 4 Reading of concentration of blood glucose level in mg/dl at different time interval 0, 15, 45, 90, 120 min.

Time (min)	Control	Glibenclamide	50b	50c	50e	50g	50a	50f	50h	50d
0	74.23± 1.23	75.45± 0.97	78.35± 2.41	76.79± 2.81	79.27± 2.05	82.43± 3.98	80.49± 3.68	83.46± 3.02	76.47± 2.58	75.72± 2.84
15	164.83 ±0.4	160.28± 2.32	171.57± 2.98	168.32± 4.53	161.23± 3.29	172.37± 4.48	173.23± 3.34	167.57± 3.43	168.23± 2.2	170.09± 4.04
45	155.63± 0.76	125.62± 3.42 **	130.56± 4.86 **	156.78± 4.02ns	138.12± 3.43 **	158.21± 3.15ns	156.85± 4.22 ns	137.21± 4.08**	139.01± 3.65*	157.26± 2.04ns
90	142.15± 0.82	110.23± 2.98 **	115.33± 2.72 **	148.21± 3.72 ns	121.31± 3.46 **	140.65± 3.56 ns	141.75± 2.98ns	124.32± 4.58 **	127.91± 2.92*	145.74± 3.02ns
120	114.61± 0.71	90.41± 2.53 *	103.67± 2.78	119.74± 3.56	109.35± 2.85	119.78± 4.02	118.44± 3.21	109.09± 3.75	112.58± 2.29	115.25± 3.47

*Values of mean ±SEM (n=6) One wary ANOVA (**, p <0.01; *, p<0.05 consider for significance) Followed by post test, Dunnett's multiple comparisiton tests*

Figure 3: Histogram showing blood glucose level in control, standard and test compounds by OGTT

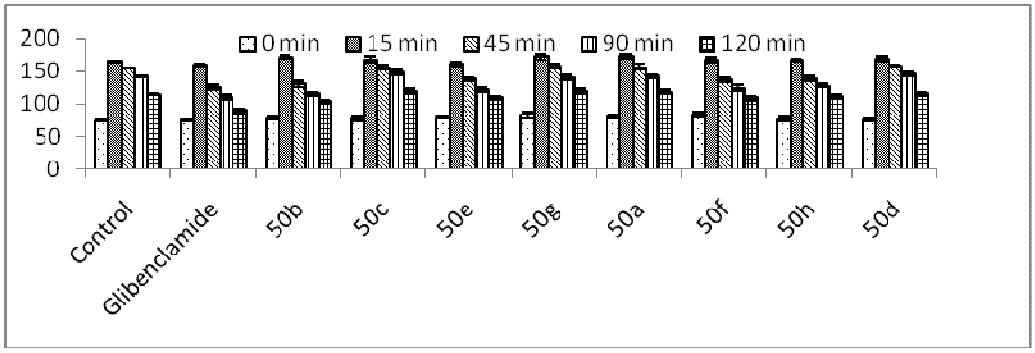
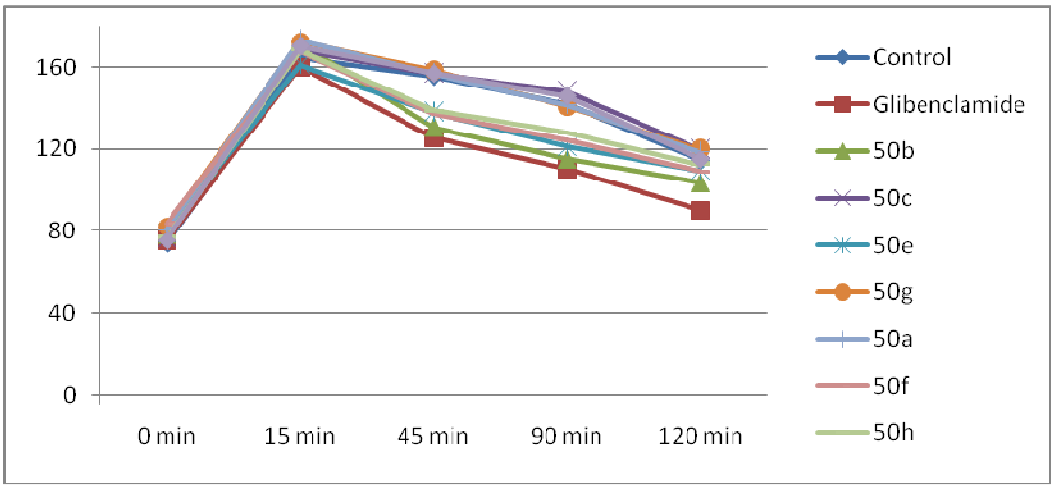


Figure 4 :Figure showing the blood glucose level by OGTT



CONCLUSION

All the synthesized compounds were subjected to OGTT to gain preliminary information regarding the ant hyperglycemic effect in normal rats. Albino wistar rats are chosen for the study. Glibenclamide was chosen as the standard. Blood samples were collected form retro orbital plexus prior to glucose administration and at 15, 45, 90 and 120 minutes after glucose loading .Blood glucose level were measured using semi auto analyzer. The data so obtained was represented in Table 4. Comparisiton of data was performed by one wary ANOVA followed by post test, Dunnett's multiple comparisiton tests (**, $p<0.01$; *, $p<0.05$ consider for significance). Compound 50b, 50e, 50f, 50h was found to be having significant glucose lowering effect as compared to control and standard. Other compounds like 50c, 50g, 50d, and 50a were found to be non significant as comparisiton to control. The synthesized compound 50b, 50e, 50f, 50h were proposed to further analysis by most relevant animal models like alloxan/streptozotocin induced diabetic animal model.

REFERENCES

[1] Http://www.who.int/mediacentre/factsheets/fs312/en /**2008**.
[2] http://www.idf.org/home/index.cfm.
[3] Http://www.who.int/diabetes/booklet_html/en/print.html.**2008.**
[4] International diabetes federation. Facts & figures: Available at: Http://www.idf.org/home/index.cfm. **2008.**
[5] UK prospective diabetes study group, intensive blood glucose Control with sulphonyl ureas or insulin compared with conventional Treatment and risk complications in patients with type 2 diabetes. *Lancet*, **1998**; 352: 837–16.
[6] Burge MR, Sood V, Sobhy TA, Rassam AG, and Schade DS, *Diabetes obesity metab*. **1999;** 1: 199–7.
[7] Amos AF, McCarty DJ, and Zimmet P, *Diabetes med.(suppl.)*, **1997**;14: s1–85.
[8] Hermenegilda MD, Rafael VM, Rolffy OA, Daniel DC, Gabriel NV, *Bioorganic & Medicinal Chemistry Letters* **2008;** 18: 2871–7.
[9] Ariful I, Afia A, Rafiqul IK, Sarowar H, Imam IW. *Pak. J. Pharm. Sci*., **2009;**.22(4),402-4.