Research Article

SYNTHESIS AND PHARMACOLOGICAL EVALUATION OF NEW QUINAZOLINONE DERIVATIVES AS ANTI DIABETIC AGENTS

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ABSTRACT: In this study, a library of new 4-[6-chloro-2-arylaminoethyl-4-oxoquinazolin-3(4H)-yl] benzoic acid (Q_1-Q_{16}) were synthesized and evaluated for their *in vivo* anti-diabetic activity. All the compounds were prepared in a multistep process involving the initial preparation of 5-chloro-N-acetyl anthranilic acid which was converted to 4-[6-chloro-2-methyl-4-oxoquinazolin-3(4H)-yl] benzoic acid. This resulted intermediate undergoes mannich base reaction in the presence of formaldehyde and different aromatic amine to produce different quinazolinone derivatives in good yield. The structures of the synthesized compounds were establishedon the basis of elemental analysis and spectroscopic studies (FTIR, ^{1}HNMR and Mass) and the purity of the compounds was determined by TLC. All the synthesized compounds were subjected to Oral Glucose Tolerance Test (OGTT) to gain preliminary information regarding the anti-diabetic activity. They were assessed for anti-diabetic actionusing glibenclamide as the standard. Among the test compounds Q_2, Q_6 and Q_{10} were significant in their anti-diabetic activity in comparison with standard while the remaining tested compounds had shown good to moderate activity.

Keywords: Alloxan monohydrate, Anthranilic acid, Anti-diabetic activity, Glibenclamide, OGTT, Quinazolinone

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INTRODUCTION:

Diabetes Mellitus (DM)is severe and very common disease affecting the populations all around the world. It has been observed that around 25% of the total world population facing this problem now days. Due to defect in metabolism of carbohydrate, diabetes may arisewhich ultimately leads to decrease the level of insulin in blood or decline the sensitivity

of target organ towards insulin[1]. Another root cause of DM is either insufficient production of insulin hormone by pancreas organ or when the body does not use it properly for smooth functioning. Blood sugar level is regulated by insulin in the body. Decrease in the level of insulin may causehyperglycaemia resulting in diminishedvarious body function or various body system such as ocular, renal, nervous, hepatic, cardiac system etc.[2,3]. Extremethirst, frequent urination, change in weight, tiredness etc. are some common sign of diabetes. Due to high percentage of mortality and morbidity, diabetes in now considered as one of the major cause of the death in the world [4,5]. Change in lipid, protein and carbohydrate metabolism and hyperglycaemia is the principal characteristics of diabetes.Several research said that various derivatives ofquinazolinone[6]were found to be highly active *in-vivo* for their anti-diabetic activity by increasing the level of insulin in the body, hencewe paid our attention towards the synthesis of various novel quinazolinone derivatives. Numerous biologically pharmaceutical active compound contain quinazolinone ring as a basic and prominent structural motif. Quinazolinone shows a wide range of potential biological actionand now present in various pharmacological active molecules[7]viz.Ispinesib/ Raltitrexed (anticancer), Albaconazole (anti-fungal), Balaglitazone (anti-diabetic) and Piriqualone (anticonvulsant). Modification in the ring and its structureprovides high degree of diversitythat help for newer development of structural compoundswith increase in its potency to greater extent and minimum level of toxicity. The titled compound quinazolinone is a six membered heterocyclic ringwith two nitrogen atoms in it which play pivotal role in medicinal chemistry and its development and used as important synthons in organic synthesis. Quinazolinone and their derivatives have shown wide range of biological activities such asanti-diabetic[8,9]. In addition, they also exhibit various activities like anti-malarial[10], analgesic[11], antioxidant[12], anticancer[13], antiviral[14], antifeedant[15], sedative-hypnotic[16], antimicrobial[17], antialgal[18], hypotensive[19] and anti-inflammatory[20].

RESULTS AND DISCUSSION:

Chemistry:

All the novel quinazolinone derivatives were synthesized, purified and separated by using column chromatography or recrystallization method. They were characterized by using Elemental analysis, FT-IR, ^{1}HNMR and Mass Spectrometric data. The integration curves fully support the orientation of protons in the analyzed compounds. Furthermore, all the compounds demonstrated the characteristic chemical shifts for the quinazolinone nucleus. Additionally, all derivativeswere analyzed by mass spectra and indicated no difference in the fragmentation pattern among the set of synthesizedseries.

Anti-diabetic Activity:

The newly synthesized compounds were screened for their anti-diabetic activity. Sixteen compounds were tested for Oral Glucose Tolerance Test for preliminary study. Among them, seven compounds Q_2, Q_5, Q_6, Q_9, Q_{10}, Q_{12} and Q_{15} were selected for further study based on OGTT. Among the test compounds Q_2, Q_6 and Q_{10} showed significance decrease in blood glucose level as 178.62 mg/dl, 197.26 mg/dl and 189.43 mg/dl respectively, which were nearby 132.56 mg/dl of the standard Glibenclamide drug used and also greater than the other quinazolinone derivatives.

Table 1: Oral Glucose Tolerance Test

Group	Com.	Dose	Blood Glucose Levels (mg/dl)				
			0 min.	30 min.	60 min.	90 min.	120 min.
I	Control (Water)	2 ml	79.15 ± 1.410	107.41 ± 2.658	104.23 ± 1.210	93.31 ± 0.917	92.55 ± 0.648
II	Standard (GBC)	10 mg	83.17 ± 1.458	103.76 ± 1.812	98.59 ± 1.350	86.89 ± 1.189	82.10 ± 0.949**
III	Q_1	10 mg	87.75 ± 1.302	154.16 ± 1.346	139.81 ± 1.195	126.30 ± 2.018	112.58 ± 1.411*
IV	Q_2	10 mg	96.07 ± 0.694	146.70 ± 1.608	98.51 ± 1.412	89.14 ± 3.121	81.69 ± 0.847**
V	Q_3	10 mg	78.35 ± 1.681	153.08 ± 1.157	136.18 ± 0.940	123.42 ± 1.678	118.47 ± 1.379*
VI	Q_4	10 mg	85.23 ± 1.890	144.28 ± 1.190	138.34 ± 1.742	121.23 ± 1.440	115.36 ± 2.213**
VII	Q_5	10 mg	91.78 ± 1.118	140.39 ± 1.160	114.61 ± 1.712	92.09 ± 1.124	87.71 ± 1.610*
VIII	Q_6	10 mg	94.42 ± 1.127	151.60 ± 1.181	104.15 ± 1.610	96.59 ± 0.781	83.78 ± 1.115**
IX	Q_7	10 mg	97.78 ± 1.052	139.92 ± 1.310	122.06 ± 1.170	104.83 ± 1.493	98.48 ± 1.717*
X	Q_8	10 mg	96.72 ± 1.248	141.57 ± 1.640	125.24 ± 1.077	113.36 ± 1.514	101.25 ± 1.539**
XI	Q_9	10 mg	93.70 ± 1.414	148.47 ± 1.329	112.75 ± 1.812	95.18 ± 2.260	84.68 ± 2.268*
XII	Q_{10}	10 mg	92.11 ± 1.829	149.52 ± 1.159	102.42 ± 1.614	90.59 ± 1.489	80.83 ± 2.132**
XIII	Q_{11}	10 mg	92.88 ± 1.719	152.42 ± 1.210	129.66 ± 1.315	111.82 ± 1.257	105.49 ± 1.419**
XIV	Q_{12}	10 mg	89.31 ± 1.610	141.23 ± 1.519	119.66 ± 1.448	99.48 ± 1.576	85.29 ± 1.639*
XV	Q_{13}	10 mg	95.40 ± 1.571	134.81 ± 1.315	127.25 ± 1.474	118.53 ± 1.520	109.17 ± 1.616*
XVI	Q_{14}	10 mg	90.54 ± 1.515	137.37 ± 1.219	121.20 ± 1.327	102.03 ± 1.680	99.57 ± 1.277*
XVII	Q_{15}	10 mg	93.49 ± 1.421	147.81 ± 1.501	113.43 ± 1.418	101.47 ± 1.207	89.71 ± 1.518**
XVIII	Q_{16}	10 mg	84.57 ± 1.910	150.32 ± 1.919	143.80 ± 1.337	129.94 ± 1.472	117.79 ± 1.369*

Values are expressed as mean ± SEM offive animals in each group. *Statistically significant ($P<0.05$), **Statistically significant ($P<0.01$)

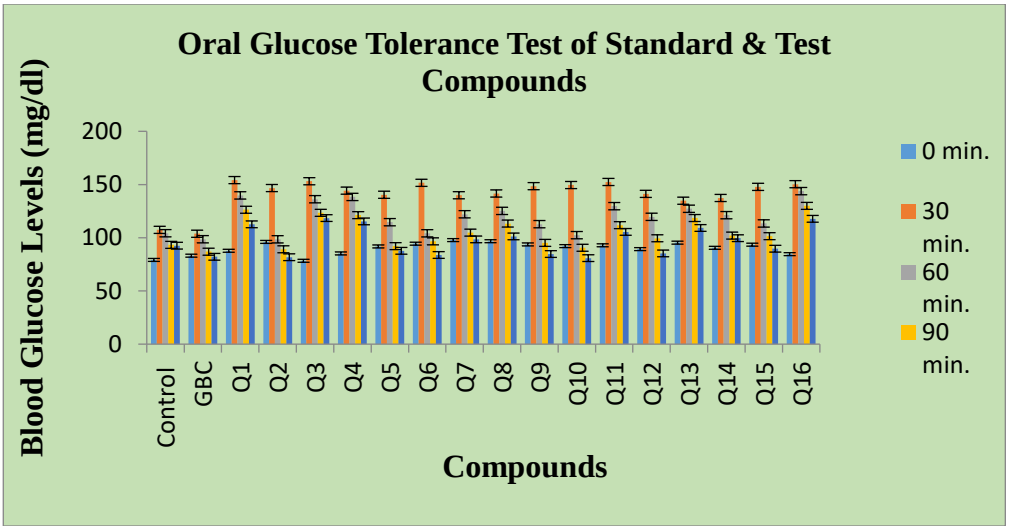


Figure1. Graphical representation of blood glucose levels by OGTT test of the quinazolinone derivatives compared to glibenclamide

Table 2: Effect on diabetic rats (Alloxan induced rats) Body Weight in Normal and Diabetic Rats (Sub Acute Study)

Compounds Q_2, Q_5, Q_6, Q_9, Q_{10}, Q_{12} and Q_{15} were chosen for the study based on Oral Glucose Tolerance Test (Table-2)

Group	Compounds	Dose	Body Weight (gm)				
			Initial Wt.	1st Day	3rd Day	7th Day	14th Day
I	Control (Water)	2 ml	190.11 ± 6.110	190.40 ± 6.110	189.25 ± 6.121	188.32 ± 6.319	188.30 ± 6.317
II	Diabetic control (Alloxan)	130 mg	184.70 ± 7.205	178.59 ± 6.650	172.34 ± 6.002	161.41 ± 6.210	149.73 ± 5.324**
III	Standard (GBC+ Alloxan)	10 mg	171.34 ± 5.612	174.88 ± 5.618	180.52 ± 5.819	183.09 ± 5.408	187.66 ± 5.121**
IV	Q_2 + Alloxan	10 mg	174.00 ± 8.109	151.28 ± 9.501	154.30 ± 7.712	157.56 ± 8.151	160.85 ± 8.120**
V	Q_5 + Alloxan	10 mg	197.45 ± 4.232	172.74 ± 5.440	170.81 ± 6.192	179.26 ± 6.510	193.52 ± 4.191*
VI	Q_6 + Alloxan	10 mg	169.83 ± 5.172	150.66 ± 4.095	149.29 ± 6.110	151.78 ± 4.752	156.43 ± 7.085**
VII	Q_9 + Alloxan	10 mg	195.72 ± 6.241	170.25 ± 6.340	172.47 ± 5.117	181.80 ± 5.419	194.75 ± 7.612*
VIII	Q_{10} + Alloxan	10 mg	210.85 ± 6.618	191.37 ± 5.550	190.70 ± 7.702	195.61 ± 6.085	201.27 ± 5.742**
IX	Q_{12} + Alloxan	10 mg	188.29 ± 7.810	172.48 ± 7.247	168.55 ± 6.547	179.49 ± 5.831	186.91 ± 6.117*
X	Q_{15} + Alloxan	10 mg	192.58 ± 4.109	178.93 ± 5.192	172.42 ± 4.317	176.06 ± 4.539	187.61 ± 6.297*

Values are expressed as mean ± SEM of five animals in each group. *Statistically significant (P<0.05), ** Statistically significant (P<0.01)

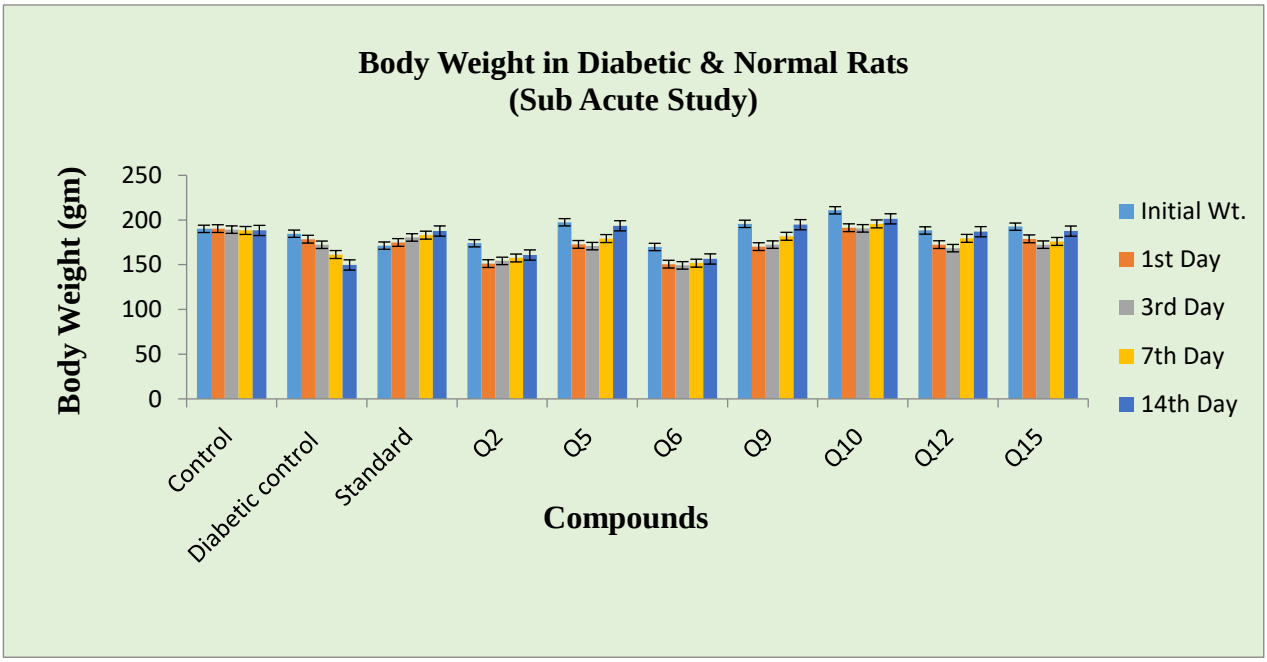


Figure 2.Graphical representation of body weights in normal and diabetic rats.

Table 3: Anti-diabetic Effect of test compounds (Sub Acute Study)

Group	Compound	Dose	Blood Glucose Level (mg/dl)			
			1st Day	3rdDay	7th Day	14th Day
I	Control (Water)	2 ml	95.42 ± 1.727	96.19 ± 1.349	98.51 ± 1.129	95.39 ± 1.356
II	Diabetic control (Alloxan)	130 mg	492.68 ± 21.290	495.27 ± 23.419	488.70 ± 19.717	483.44 ± 22.297**
III	Standard (GBC+ Alloxan)	10 mg	413.18 ± 18.170	368.46 ± 19.564	251.35 ± 20.027	132.56 ± 18.225**
IV	Q_2 + Alloxan	10 mg	479.06 ± 22.215	388.55 ± 24.721	246.72 ± 23.194	178.62 ± 23.107**
V	Q_5 + Alloxan	10 mg	501.52 ± 20.192	390.33 ± 17.692	318.21 ± 16.510	237.18 ± 17.517*
VI	Q_6 + Alloxan	10 mg	452.79 ± 19.272	382.16 ± 18.523	291.60 ± 17.619	197.26 ± 13.279**
VII	Q_9 + Alloxan	10 mg	518.27 ± 23.117	419.47 ± 24.781	309.82 ± 21.210	224.08 ± 23.171*
VIII	Q_{10} + Alloxan	10 mg	443.12 ± 21.819	327.62 ± 21.616	231.15 ± 22.450	189.43 ± 23.770**
IX	Q_{12} + Alloxan	10 mg	506.84 ± 18.181	431.64 ± 17.169	358.56 ± 15.249	265.93 ± 15.663*
X	Q_{15} +	10 mg	511.18 ±	477.36 ±	346.20 ±	248.71 ±

	Alloxan		13.179	14.606	14.832	12.190*

Values are expressed as mean ± SEM of five animals in each group. * Statistically significant (P<0.05), ** Statistically
significant (P<0.01)

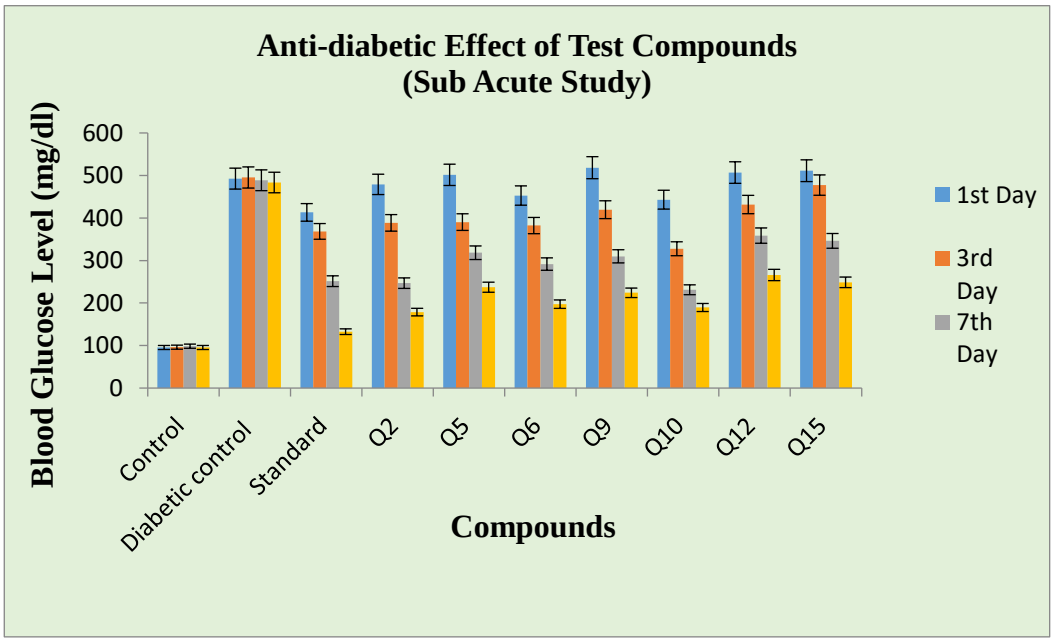


Figure 3: Graphical representation of blood glucose levels in normal and diabetic rats

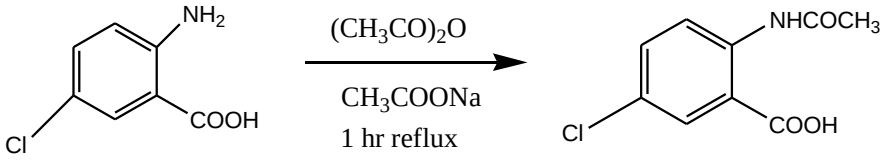
EXPERIMENTAL:

Materials and Methods:

All the chemicals used in synthesis of the intermediates and final derivatives were of A.R grade and procured from the Merck and LOBA chemicals. All the synthesized quinazolinone derivatives were characterized by melting point determination using Veergo digital melting point apparatus in open capillary tubes and were uncorrected.

IR spectra were recorded using Perkin Elmer FTIR spectrophotometer using KBr pellets techniques and ^{1}HNMR spectra of the synthesized compounds in deuteriated DMSO were recorded on BRUKER ADVANCE II 400MHz. NMR Spectrometer instrument using TMS as the internal standard. Mass spectra were recorded using LC-MSD-Tranp-SL2010A SHIMADZU using DMSO as solvent. TLC was done by using silica gel GF_{254}coated platesof 0.25 mm thickness. Ethyl acetate, petroleum ether, chloroform (0.6:0.8:8.6) were used as solvent system and iodine vapours as visualizing agent.

Scheme of Synthesis:



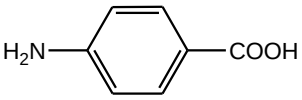
5-chloro anthranilic acid

5-chloro-N-acetyl anthranilic acid

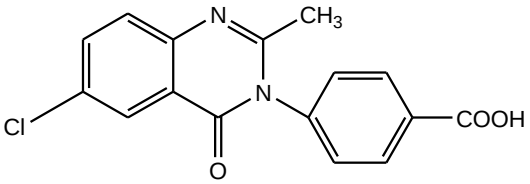
Cyclization

6 hrs reflux

i) P_2O_5 / Glacial acetic acid

ii) 

para-amino benzoic acid



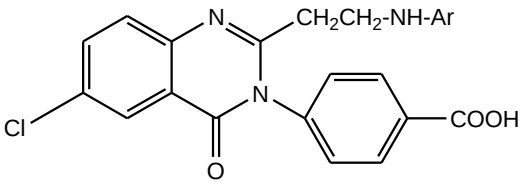
4-[6-chloro-2-methyl-4-oxoquinazolin-3(4H)-yl]benzoic acid

Mannich Reaction

4 hrs reflux

i) HCHO

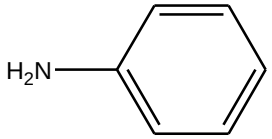
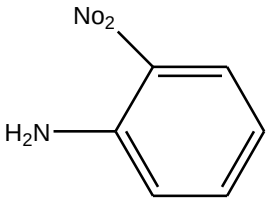
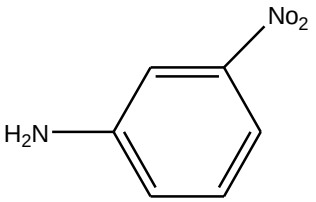
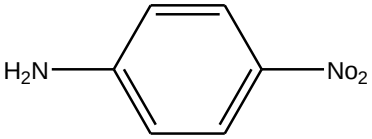
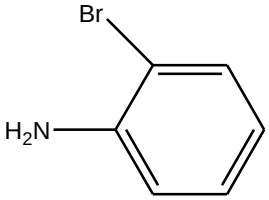
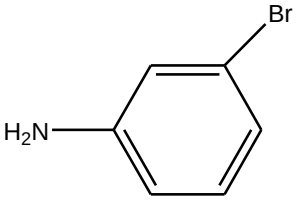
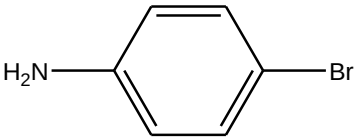
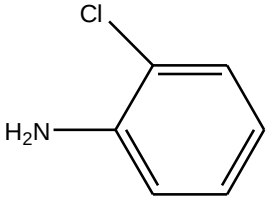
ii) $ArNH_2/Ar_2NH$

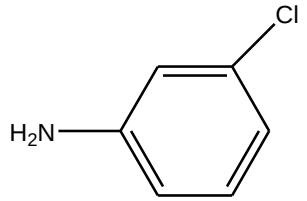
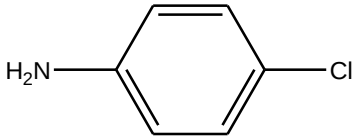
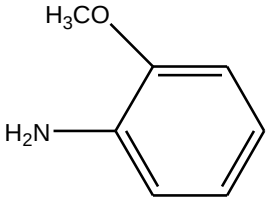
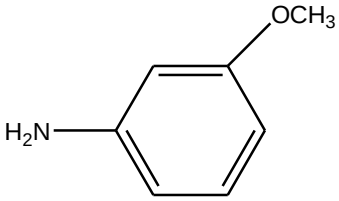
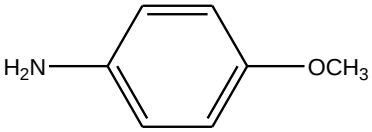
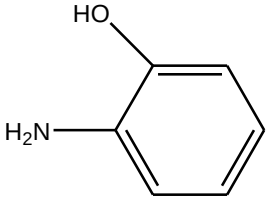
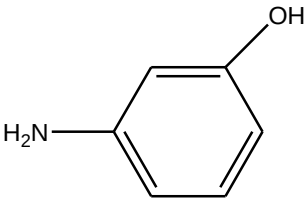
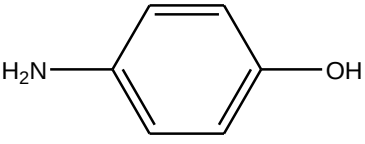


4-[6-chloro-2-arylaminoethyl-4-oxoquinazolin-3(4H)-yl]benzoic acid

Ar = *DIFFERENT AROMATIC AMINES*

Table 4: LIST OF VARIOUS AROMATIC AMINES

S. No.	Compounds Code	Substituted Aromatic Amine (Ar)	Structure of Aromatic Amine (Ar)
1	Q_1	Aniline	
2	Q_2	o-nitro aniline	
3	Q_3	m-nitro aniline	
4	Q_4	p-nitro aniline	
5	Q_5	o-bromo aniline	
6	Q_6	m-bromo aniline	
7	Q_7	p-bromo aniline	
8	Q_8	o-chloro aniline	

9	Q_9	m-chloro aniline	
10	Q_{10}	p-chloro aniline	
11	Q_{11}	o-methoxy aniline	
12	Q_{12}	m-methoxy aniline	
13	Q_{13}	p-methoxy aniline	
14	Q_{14}	o-hydroxy aniline	
15	Q_{15}	m-hydroxy aniline	
16	Q_{16}	p-hydroxy aniline	

The Experimental Work Comprises in Three Steps.

Step-I: Synthesis of 5-chloro-N-acetyl anthranilic acid from 5-chloro anthranilic acid.

Step-II: Synthesis of 4-[6-chloro-2-methyl-4-oxo quinazolin-3(4H)-yl] benzoic acid.

Step-III: Synthesis of various derivatives of quinazolinone by mannich reaction.

Step-I: General procedure for the synthesis of 5-chloro-N-acetyl anthranilic acid from 5-chloro anthranilic acid (Intermediate-I):

5-Chloro anthranilic acid (0.02 moles) was mixed with an equimolar quantities of anhydrous sodium acetate (0.03moles) and acetic anhydride (0.04 moles in slight excess) and refluxed on sand bath under anhydrous condition for 1 hr. Then the reaction mixture was poured in ice cold water and the crude product was filtered and dried. The dried crude product was recrystallized from ethanol. Yield: 81.34% M.P.: 188-190^{o}C.

Step-II: General procedure for the synthesis of 4-[6-chloro-2-methyl-4-oxo quinazolin-3(4H)-yl] benzoic acid (Intermediate-II):

5-Chloro-N-acetyl anthranilic acid (0.01 moles) was added to a mixture of 4-amino benzoic acid (0.02 moles), Phosphorus pentoxide (0.03 moles) and Glacial acetic acid (15 ml) and the mixture was refluxed under anhydrous condition for 6 hrs.Then the reaction mixture was poured into 10% Sodium bicarbonate solution (50 ml) and crude product was filtered and dried. The dried crude product was recrystallized from ethanol. Yield: 76.67% M.P.: 220-222^{o}C.

Step-III: General procedure for the synthesis of various derivatives of quinazolinonebymannich reaction(Q_1-Q_{16})

4-[6-chloro-2-arylaminoethyl-4-oxoquinazolin-3(4H)-yl] benzoic acid:

A mixture of 4-[6-chloro-2-methyl-4-oxoquinazolin-3(4H)-yl] benzoic acid (0.01 mole), various aromatic amines (0.02 mole) and formaldehyde (0.02 mole) were taken in methanol (80 ml) and the reaction mixture was refluxed for 4 hrs. The completion of reaction was monitored by TLC. The excess of the solvent was distilled off and the residue was recrystallized from acetone to give final product.

Q_1: 4-(6-chloro-4-oxo-2-(2-(phenylamino)ethyl)quinazolin-3(4H)-yl)benzoic acid:

Dark brown colored solid, Molecular formula: $C_{23}H_{18}ClN_3O_3$, Molecular weight: 419.86, Yield: 69.22%, M.P.: 176-178^{o}C, R_f value: 0.79, **FT-IR (KBr, cm^{-1}):** 3407.07 (N-H Str.), 2905.12 (C-H Str.), 1609.91 (C=C Str.), 1711.94 (C=O Str.), 1250.04 (C-N Str.), 734.56 (Ar C-H Bend.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):** 1.55 (t, 2H, CH_2), 3.22 (q, 2H, CH_2), 4.13 (t, 1H, NH), 6.38-8.10 (m, 12H,Ar H), 11.10 (s, 1H, COOH).**Mass Spectra: m/z:** 421.67 (M^{+2}). **Elemental Analysis, % found(% required):**C 65.64 (65.79);H 4.28

(4.32);N 9.93 (10.01); O 11.32 (11.43);

Cl 8.38 (8.44).

Q_2: 4-(6-chloro-2-(2-(2-nitrophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid: Yellowish brown colored solid, Molecular formula: $C_{23}H_{17}ClN_4O_5$, Molecular weight: 464.86, Yield: 68.32%, M.P.: 152-154^oC, $R_f$value: 0.79,**FT-IR (KBr, cm^{-1}):** 3434.67 (N-H Str.), 2991.28 (C-H Str.), 1629.41 (C=C Str.), 1701.03 (C=O Str.), 1254.04 (C-N Str.), 743.35 (Ar C-H Bend.), 1497.09 (Ar N=O Str.). **^{1}H-NMR (400 MHz, DMSO, δ ppm**) 1.57 (t, 2H, CH_2), 3.31 (q, 2H, CH_2), 4.20 (t, 1H, NH), 6.70-8.14 (m, 11H, Ar H), 11.12 (s, 1H, COOH).**Mass Spectra: m/z:** 466.43 (M^{+2}). **Elemental Analysis, % found (% required):**C 59.34 (59.43);H 3.65 (3.69);N 11.96 (12.05);

O 17.15 (17.21); Cl 7.55 (7.63).

Q_3: 4-(6-chloro-2-(2-(3-nitrophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid: Creamish yellow colored solid, Molecular formula: $C_{23}H_{17}ClN_4O_5$, Molecular weight: 464.86, Yield: 67.22%, M.P.: 168-170^oC,R_f value: 0.80, **FT-IR (KBr, cm^{-1}):** 3396.21 (N-H Str.), 2895.47 (C-H Str.), 1599.85 (C=C Str.), 1704.19 (C=O Str.), 1222.75 (C-N Str.), 742.59 (ArC-H Bend.), 1452.39 (Ar N=O Str.).**^{1}H-NMR (400 MHz, DMSO, δ ppm**):1.58 (t, 2H, CH_2), 3.29 (q, 2H, CH_2), 4.17 (t, 1H, NH), 6.75-8.12 (m, 11H, Ar H), 11.00 (s, 1H, COOH). **Mass Spectra: m/z:** 466.57 (M^{+2}). **Elemental Analysis, % found(% required):**C 59.35 (59.43); H 3.51 (3.69); N 12.08 (12.05);

O 17.12 (17.21); Cl 7.52 (7.63).

Q_4: 4-(6-chloro-2-(2-(4-nitrophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Pale yellow colored solid, Molecular formula: $C_{23}H_{17}ClN_4O_5$, Molecular weight: 464.86, Yield: 67.77%, M.P.: 178-180^oC, R_f value: 0.76, **FT-IR(KBr, cm^{-1}):** 3434.65 (N-H Str.), 2917.59 (C-H Str.), 1657.18 (C=C Str.), 1754.78 (C=O Str.), 1259.94 (C-N Str.), 796.20 (Ar C-H Bend.), 1470.73 (Ar N=O Str.). **^{1}H-NMR (400 MHz, DMSO,δ ppm):**1.61 (t, 2H, CH_2), 3.15 (q, 2H, CH_2), 4.11 (t, 1H, NH), 6.65-8.12 (m, 11H, Ar H), 11.15 (s, 1H, COOH).**Elemental analysis, % found (% required):**C59.37 (59.43); H3.61 (3.69); N 11.95 (12.05); O 17.13 (17.21); Cl 7.60 (7.63).

Q_5: 4-(2-(2-(2-bromophenylamino)ethyl)-6-chloro-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Pale red colored solid, Molecular formula:$C_{23}H_{17}BrClN_3O_3$, Molecular weight: 498.76, Yield: 73.24%, M.P.: 155-157^oC, $R_f$value: 0.76, **FT-IR (KBr, cm^{-1}):** 3363.76 (N-H Str.), 2898.37 (C-H Str.), 1599.58 (C=C Str.), 1679.43 (C=OStr.), 1258.02 (C-N Str.), 768.37

(ArC-H Bend.), 678.46 (Ar C-Br Bend.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.63 (t, 2H, CH_2), 3.12 (q, 2H, CH_2), 4.14 (t, 1H, NH), 6.34-8.11 (m, 11H, Ar H), 11.10 (s, 1H, COOH). **Mass Spectra: m/z:** 500.07 (M^{+2}). **Elemental Analysis, % found (% required):**C 55.32 (55.39); H3.40 (3.44); N 8.35 (8.42);
O 9.57 (9.62); Cl 7.07 (7.11); Br 15.99 (16.02).

Q_6: 4-(2-(2-(3-bromophenylamino)ethyl)-6-chloro-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Light red colored solid, Molecular formula: $C_{23}H_{17}BrClN_3O_3$, Molecular weight: 498.76, Yield: 72.84%, M.P.: 158-160^{o}C, $R_f$value: 0.74, **FT-IR (KBr, cm^{-1}):** 3320.88 (N-H Str.), 2809.58 (C-H Str.), 1589.43 (C=C Str.), 1666.88 (C=O Str.), 1258.46 (C-N Str.), 718.27 (ArC-H Bend.), 650.43 (Ar C-Br Bend.). **^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.64 (t, 2H, CH_2), 3.10 (q, 2H, CH_2), 4.16 (t, 1H, NH), 6.37-8.13 (m, 11H, Ar H), 11.08 (s, 1H, COOH). **Mass Spectra: m/z:** 500.04 (M^{+2}). **Elemental Analysis, % found(% required):**C 55.32 (55.39); H 3.38 (3.44); N 8.36 (8.42);
O 9.57 (9.62); Cl 7.10 (7.11); Br 15.98 (16.02).

Q_7: 4-(2-(2-(4-bromophenylamino)ethyl)-6-chloro-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Greyish red colored solid, Molecular formula: $C_{23}H_{17}BrClN_3O_3$, Molecular weight: 498.76, Yield: 70.24%, M.P.: 160-162^{o}C, $R_f$value: 0.70, **FT-IR (KBr, cm^{-1}):** 3334.67 (N-H Str.), 2849.31 (C-H Str.), 1597.36(C=C Str.), 1693.03 (C=O Str.), 1255.20 (C-N Str.), 717.26 (ArC-H Bend.), 637.77 (Ar C-Br Bend.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):** 1.57 (t, 2H, CH_2), 3.14 (q, 2H, CH_2), 3.94 (t, 1H, NH), 6.29-8.10 (m, 11H, Ar H), 11.05 (s, 1H, COOH). **Elemental Analysis, % found(% required):**C 55.36 (55.39); H 3.41 (3.44); N 8.34 (8.42); O 9.59 (9.62); Cl 7.02(7.11); Br 15.94 (16.02).

Q_8: 4-(6-chloro-2-(2-(2-chlorophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Dark Browncoloredsolid, Molecular formula: $C_{23}H_{17}Cl_2N_3O_3$, Molecular weight: 454.31, Yield: 69.30%, M.P.: 204-206^{o}C, R_f value: 0.71, **FT-IR (KBr, cm^{-1}):** 3371.64 (N-H Str.), 2863.85 (C-H Str.), 1572.74(C=C Str.), 1711.10 (C=O Str.), 1259.33 (C-N Str.), 713.32 (ArC-H Bend.), 654.23 (Ar C-Cl Bend.).**^{1}H-NMR (400MHz, DMSO, δ ppm):**1.59 (t, 2H, CH_2), 2.98 (q, 2H, CH_2), 4.03 (t, 1H, NH), 6.37-8.12 (m, 11H, Ar H), 11.02 (s, 1H, COOH). **Mass Spectra: m/z:** 456.39 (M^{+2}). **Elemental Analysis, % found (% required):**C 60.77 (60.81); H 3.72 (3.77); N 9.20 (9.25);
O 10.51 (10.57); Cl 15.58 (15.61).

Q_9: 4-(6-chloro-2-(2-(3-chlorophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Pale Brown colored solid, Molecular formula: $C_{23}H_{17}Cl_2N_3O_3$, Molecular weight: 454.31, Yield: 67.84%, M.P.: 210-212^oC, $R_f$value: 0.78, **FT-IR(KBr, cm^{-1}):** 3394.98 (N-H Str.), 2858.37 (C-H Str.), 1504.84 (C=C Str.), 1724.98 (C=O Str.), 1209.33 (C-N Str.), 710.11 (ArC-H Bend.), 673.29 (Ar C-Cl Bend.). **^{1}H-NMR (400 MHz, DMSO,δ ppm):**1.62 (t, 2H, CH_2), 3.11 (q, 2H, CH_2), 4.08 (t, 1H, NH), 6.30-8.07 (m, 11H, Ar H), 11.01 (s, 1H, COOH). **Elemental Analysis, % found(% required):**C 60.77 (60.81); H3.71 (3.77); N 9.22 (9.25); O 10.54 (10.57); Cl 15.58 (15.61).

Q_{10}: 4-(6-chloro-2-(2-(4-chlorophenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Creamish Brown colored solid, Molecular formula: $C_{23}H_{17}Cl_2N_3O_3$, Molecular weight: 454.31, Yield: 66.67%, M.P.: 209-211^oC, $R_f$value: 0.69, **FT-IR (KBr, cm^{-1}):** 3375.68 (N-H Str.), 2719.68 (C-H Str.), 1531.28 (C=C Str.), 1717.59 (C=O Str.), 1207.04 (C-N Str.), 761.10 (ArC-H Bend.), 640.39 (Ar C-Cl Bend.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.60 (t, 2H, CH_2), 3.17 (q, 2H, CH_2), 4.10 (t, 1H, NH), 6.35-8.11 (m, 11H,Ar H), 10.89 (s, 1H, COOH). **Mass Spectra: m/z:** 456.69 (M^{+2}). **Elemental Analysis, % found (% required):**C 60.78 (60.81); H 3.74 (3.77); N 9.19 (9.25); O 10.50 (10.57); Cl 15.58 (15.61).

Q_{11}: 4-(6-chloro-2-(2-(2-methoxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Yellowish White colored solid, Molecular formula: $C_{24}H_{20}ClN_3O_4$, Molecular weight: 449.89, Yield: 64.54%, M.P.: 147-149^oC, R_f value: 0.65, **FT-IR (KBr, cm^{-1}):** 3369.95 (N-H Str.), 2809.33 (C-H Str.), 1517.09(C=C Str.), 1694.15 (C=O Str.), 1217.93 (C-N Str.), 710.77 (ArC-H Bend.). **^{1}H-NMR (400 MHz, DMSO, δ ppm):** 1.61 (t, 2H, CH_2), 3.22 (q, 2H, CH_2), 4.16 (t, 1H, NH), 3.71 (s, 3H, OCH_3), 6.31-8.09 (m, 11H,Ar H), 11.04 (s, 1H, COOH). **Mass Spectra: m/z:** 451.27 (M^{+2}). **Elemental Analysis, % found (% required):**C 63.98 (64.07); H 4.45 (4.48); N 9.30 (9.34);
O 14.19 (14.23); Cl 7.84 (7.88).

Q_{12}: 4-(6-chloro-2-(2-(3-methoxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

CreamishWhite colored solid, Molecular formula: $C_{24}H_{20}ClN_3O_4$, Molecular weight: 449.89, Yield: 60.53%, M.P.: 152-154^oC, $R_f$value: 0.68, **FT-IR (KBr, cm^{-1}):** 3396.75 (N-H Str.), 2898.47 (C-H Str.), 1531.07(C=C Str.), 1704.58 (C=O Str.), 1239.75 (C-N Str.), 719.43 (ArC-H Bend.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.58 (t, 2H, CH_2), 3.11 (q, 2H, CH_2),

4.13 (t, 1H, NH), 3.75 (s, 3H, OCH_3), 5.91-8.0 (m, 11H,Ar H), 11.01 (s, 1H, COOH). **Elemental analysis, % found(% required):**C 63.99 (64.07); H 4.45 (4.48); N 9.32(9.34); O 14.19 (14.23); Cl 7.81 (7.88).

Q_{13}: 4-(6-chloro-2-(2-(4-methoxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

White Brown colored solid, Molecular formula: $C_{24}H_{20}ClN_3O_4$, Molecular weight: 449.89, Yield: 70.79%, M.P.: 138-140^oC, $R_f$value: 0.66, **FT-IR (KBr, cm^{-1}):** 3421.07 (N-H Str.), 2918.12 (C-H Str.), 1546.13(C=C Str.), 1719.41(C=O Str.), 1208.08 (C-N Str.), 735.04 (ArC-H Bend.).**^{1}H-NMR (400 MHz, DMSO,δ ppm):**1.64 (t, 2H, CH_2), 3.14 (q, 2H, CH_2), 4.07 (t, 1H, NH), 3.72 (s, 3H, OCH_3), 6.30-8.03 (m, 11H, Ar H), 10.86 (s, 1H, COOH). **Mass Spectra: m/z:** 451.35 (M^{+2}). **Elemental Analysis, % found (% required):**C 64.01 (64.07); H 4.46 (4.48);N 9.30(9.34); O 14.18 (14.23); Cl 7.80 (7.88).

Q_{14}: 4-(6-chloro-2-(2-(2-hydroxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Greyish Black colored solid, Molecular formula: $C_{23}H_{18}ClN_3O_4$, Molecular weight: 435.86, Yield: 72.11%, M.P.: 133-135^oC, $R_f$value: 0.71, **FT-IR (KBr, cm^{-1}):** 3478.22 (N-H Str.), 2934.89 (C-H Str.), 1530.86(C=C Str.), 1643.50 (C=O Str.), 1209.79 (C-N Str.), 737.35 (ArC-H Bend.), 3446.18 (Ar C-OH Str.). **^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.69 (t, 2H, CH_2), 3.25 (q, 2H, CH_2), 4.01 (t, 1H, NH), 5.10 (s, 1H, OH), 6.24-8.12 (m, 11H, Ar H), 11.12 (s, 1H, COOH). Mass **Spectra: m/z:** 437.11 (M^{+2}). **Elemental Analysis, % found (% required):**C 63.35 (63.38); H 4.10 (4.16);
N 9.59(9.64); O 14.66 (14.68);Cl 8.09 (8.13).

Q_{15}: 4-(6-chloro-2-(2-(3-hydroxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Yellowish Black colored solid, Molecular formula: $C_{23}H_{18}ClN_3O_4$, Molecular weight: 435.86, Yield: 74.38%, M.P.: 138-140^oC, $R_f$value: 0.74, **FT-IR (KBr, cm^{-1}):** 3477.38 (N-H Str.), 2979.13 (C-H Str.), 1531.84(C=C Str.), 1622.98 (C=O Str.), 1207.57 (C-N Str.), 762.11 (Ar C-H Bend.), 3446.93 (ArC-OH Str.).**^{1}H-NMR (400 MHz, DMSO, δ ppm):**1.65 (t, 2H, CH_2), 3.19 (q, 2H, CH_2), 4.04 (t, 1H, NH), 5.07 (s, 1H, OH), 5.89-8.14 (m, 11H, Ar H), 11.03 (s, 1H, COOH). **Elemental analysis, % found(% required):**C 63.33 (63.38); H 4.11 (4.16); N 9.60 (9.64); O 14.63 (14.68); Cl 8.08 (8.13).

Q_{16}: 4-(6-chloro-2-(2-(4-hydroxyphenylamino)ethyl)-4-oxoquinazolin-3(4H)-yl) benzoic acid:

Black Red colored solid, Molecular formula: $C_{23}H_{18}ClN_3O_4$, Molecular weight: 435.86,

Yield: 70.25%, M.P.: 144-146^{o}C, $R_f$value: 0.69, **FT-IR (KBr, cm^{-1}):** 3377.38 (N-H Str.), 2979.49 (C-H Str.), 1572.38(C=C Str.), 1617.87 (C=O Str.), 1249.85 (C-N Str.), 733.59 (Ar C-H Bend.), 3315.87 (ArC-OH Str.). **^{1}H-NMR (400 MHz, DMSO,δ ppm):**1.63 (t, 2H, CH_2), 3.18 (q, 2H, CH_2), 4.11 (t, 1H, NH), 5.02 (s, 1H, OH), 6.25-8.11 (m, 11H, Ar H), 11.10 (s, 1H, COOH). **Elemental Analysis, % found(% required):**C 63.31 (63.38); H 4.11 (4.16); N 9.60 (9.64); O 14.66 (14.68);
Cl 8.07 (8.13).

Biological Study:

Evaluation of Anti-diabetic Activity[21, 22, 23]**:**

Experimental animals:

Wistar albino adult rats weighing (150-200 g) were used for studying the anti-diabetic activity. The animals (five per cage) were maintained under standard laboratory conditions (light period of 12 hrs/day, temperature 27±2 ^{0}C with relative humidity of 45-55%). They were fed with standard animal feed and water *ad libitum*. The experimental procedures were carried out in strict compliance with the Institutional Animals Ethics Committee. All experiments were performed in the morning according to the guidelines for the care of laboratory animals.

Effect of Oral Glucose Tolerance Test (OGTT)

16 test compounds were used for this test. The animals were divided into 18 groups (n = 5)

Group-I: Rats served as normal control and received water.

Group-II: Rats served as standard and received glibenclamide (10 mg/kg)

Group-III-Group-XVIII: Rats were administered (10 mg/kg body weight) orally with test compounds.

For Oral Glucose Tolerance Test, overnight fasted animals were loaded with glucose (2 g/kg, i.p.), 60 min. after the administration of test compounds and the blood samples were collected on 0, 30, 60, 90, 120 minutes time interval and the blood glucose levels were determined by making use of SUGAR SCAN Glucometer (Thyrocare).

Estimation of glucose:

The blood sample was collected in the tail portion of the albino rats. Wash the rat tail with warm, soapy water. Rinse well and dry them thoroughly. Tail was also clean with alcohol pad to dry before testing. Prepare the lancing device. Take one test strip out of the test strip vial replace the vial cap immediately and close it tightly. Insert this test strip (with the black bars

facing up) into the test strip of the meter. The meter turns on automatically and the code number appears the test strip vial or can press the strip symbol which will instruct after to insert a test strip. Insert test strip within 1 minute, then the meter will display the code number. Place the lancing device in rat's tail and press the trigger, gently squeeze the rat tail until get a drop of blood. The blood sample will be drawn into the test strip automatically, hear a beet letting know the test has begun. The blood glucose level display on the monitor.(Tail tipping method)

Experimental Induction of Diabetes:

The animals (male rats) were fasted for 24 hrs and diabetes was induced by a single intraperitoneal injection of a freshly prepared solution of Alloxan monohydrate (130 mg/kg) in ice cold 0.9% saline (NaCl) solution. The animals were given 2 ml of 5% dextrose solution using oragastric tube immediately after induction to overcome the drug induced hypoglycaemia. Seventy two hours later, rats with blood glucose levels (BGLs) above 200 mg/dl were considered diabetic and selected for the experiments.

Grouping of animals: The rats were divided into ten groups of five (n = 5) each randomly.

Group-I: Rats served as normal control and received 5% gum acacia.

Group-II: Diabetic rats received 5% gum acacia served as diabetic control.

Group-III: Diabetic rats served as standard and received glibenclamide (10 mg/kg body weight)

Group-IV- Group-X: Rats were administered (10 mg/kg body weight) orally with test compounds.

CONCLUSION:

The main focus of this research work was to synthesize novel series of quinazolinonederivatives, purify, characterize and evaluate their anti-diabetic activity. From the results, it can be concluded that the modified quinazolinone show significant biological evaluation as anti-diabetic agents. However, further evaluation of quinazolinone will be undertaken, concerning the structural arrangements inring for anti-diabeticactivity.

Abbreviations:ATP, Adenosine Tri Phosphate; DM, Diabetes Mellitus; DMSO, Dimethyl Sulfoxide;

TMS, TetramethylSilane; OGTT, Oral Glucose Tolerance Test; BGL, Blood Glucose Level

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CONFLICT OF INTEREST:

The authors declared no conflict of interest.

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