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Short communication

Synthesis and blood glucose lowering effect of novel pyridazinone substituted benzenesulfonylurea derivatives

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ABSTRACT

Fifteen novel pyridazinone substituted benzenesulfonylurea derivatives (**3a**–**o**) were synthesized from corresponding sulfonamides derivatives via novel carbamates (**2a**–**e**). These were characterized by elemental analysis and various spectroscopic methods viz. IR, ^{1}H NMR, ^{13}C NMR and MS. Blood sugar lowering effect of thirteen (**3a**–**c**, **3e**, **3g**–**o**) sulfonylurea derivatives at the dose of 20 mg/kg (p.o.) were assessed using glucose tolerance test in normal and NIDDM (n2-STZ) rat models. All compounds except **3c**, **3e** and **3o** almost completely prevented the rise of blood glucose of NIDDM rats as compared with NIDDM control. While compounds **3c** and **3o** showed more than 50% prevention in the rise of blood glucose levels. In glucose-fed normal rats these compounds at the same dose except **3e** significantly prevented the rise of blood glucose (more than 50%) when compared with control of glucose-fed normal rats. From the results, novel compounds (**3a**–**c**, **3g**–**n**) exhibited considerably potent blood glucose lowering activity and may be used as lead compounds for developing new antidiabetic drugs. Some structure–activity relationship was observed while varying nature of 'Ar' and 'R'.

1. Introduction

Diabetes mellitus (DM) is one of the most daunting challenges posed by chronic diseases resulting from insulin deficiency or insulin resistance. Recent data show that approximately 135 million people suffer from diabetes mellitus worldwide, and that this number will rise to almost 300 million by the year 2025. While the rise will be of the order of 45% in developed countries, it will be almost 200% in developing countries. India has 35 million diabetics. As per WHO data the number would touch 50 million in 2020. Generally DM is classified as Type I Insulin Dependent Diabetes Mellitus (IDDM) caused by low or insufficient secretion of insulin by pancreas and Type II Non-Insulin Dependent Diabetes Mellitus (NIDDM) caused due to insufficient utilization of insulin [1,2]. NIDDM is the most common form of diabetes constituting nearly 90% of the diabetic population in any country.

Chronic diabetes is accompanied by complications such as neuropathy, nephropathy, cataracts and retinopathy, which practically are not controlled by insulin. These complications are considerably caused by accumulation of sorbitol, which is produced from glucose by aldose reductase (AR) in polyol pathway. AR converts glucose to sorbitol only at high glucose levels in plasma and tissue in diabetes [3–5]. The result of clinical trials justifies the importance of an improved glycemic control in diabetic patients in order to prevent or at least to delay long-term complications. The difficulty in obtaining normalization of blood glucose values has underlined the importance of the search for new and effective aldose reductase inhibitors (ARIs) to control the consequences of elevated glucose levels and thereby delaying the onset and retarding the progression of diabetic complications. There is need to develop antidiabetic agents provided with ARI.

Over the last forty years oral therapy for type II has focused on sulfonylureas and biguanides [6,7]. Sulfonylurea (SU) drugs improve glucose levels by stimulating insulin secretion by the pancreatic β-cell. Recently compounds containing pyridazine nucleus Chart 1 have been reported as AR inhibitors [8–10].

Therefore, it has been considered worthwhile to attach pyridazinone ring to benzenesulfonylurea derivatives. In the present study fifteen novel pyridazinone substituted benzenesulfonylurea derivatives were synthesized from corresponding sulfonamide derivatives via novel carbamates. These were characterized by elemental analysis and various spectroscopic methods viz. IR, ^{1}H NMR, ^{13}C NMR and MS. Oral antihyperglycemic efficacy of thirteen out of fifteen sulfonylurea derivatives were assessed using an oral glucose tolerance test in normal and NIDDM rat model.

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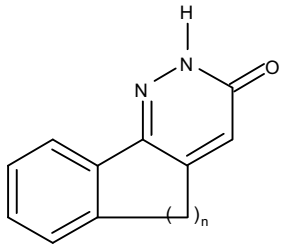


1-3
1: n = 1
2: n = 2
3: n = 3

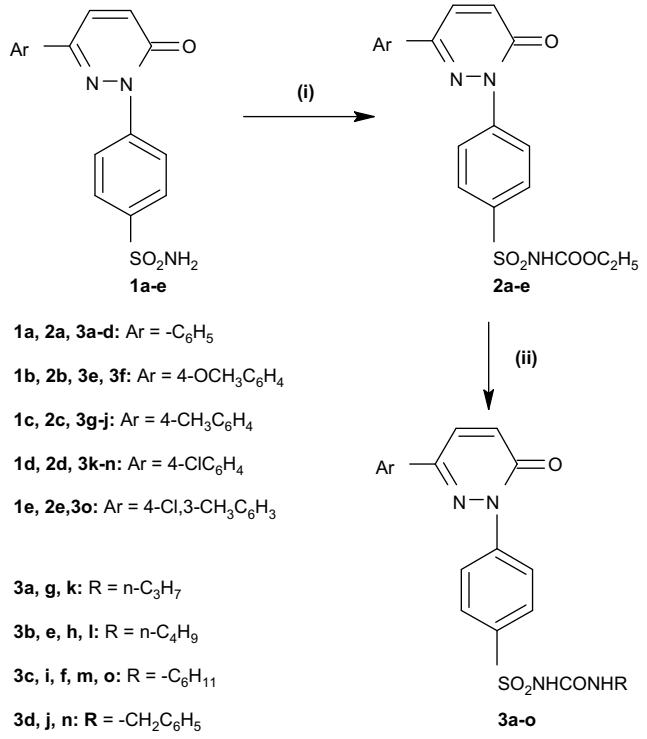
Chart 1. Structure of some compounds possessing pyridazinone nucleus reported as AR inhibitors.

2. Results and discussion

2.1. Chemistry

The synthetic route used to synthesize title compounds (**3a**–**o**) is outlined in Scheme 1. The pyridazinone substituted benzenesulfonamide derivatives (**1a**–**e**) synthesized through reported method [11] were converted to corresponding novel carbamates (**2a**–**e**) by refluxing with ethyl chloroformate in dry acetone containing anhydrous K_2CO_3. The resulting carbamates were subsequently condensed with desired primary amines to give novel pyridazinone substituted benzenesulfonylurea derivatives (**3a**–**o**).

The structures of sulfonylurea derivatives (**3a**–**o**) were determined on the basis of elemental analysis and various spectroscopic methods such as IR, ^{1}H NMR, ^{13}C NMR and MS, while carbamates (**2a**–**e**) were characterized by IR, ^{1}H NMR, MS and elemental analysis. Elemental analysis (C, H, N and S) data were within ± 0.4% of the theoretical values.



1a, 2a, 3a-d: Ar = -C_6H_5
1b, 2b, 3e, 3f: Ar = 4-$OCH_3C_6H_4$
1c, 2c, 3g-j: Ar = 4-$CH_3C_6H_4$
1d, 2d, 3k-n: Ar = 4-ClC_6H_4
1e, 2e,3o: Ar = 4-Cl,3-$CH_3C_6H_3$

3a, g, k: R = n-C_3H_7
3b, e, h, l: R = n-C_4H_9
3c, i, f, m, o: R = -C_6H_{11}
3d, j, n: R = -$CH_2C_6H_5$

Scheme 1. Reagent and conditions: (i) $ClCOOC_2H_5$, K_2CO_3, dry acetone, REFLUX 18–24H. (ii) RNH_2, toluene, reflux **3**–**4h**.

In the IR spectra of **3a**–**o** five bands characteristic of sulfonylurea moiety out of which three bands for NH (3390–3305 cm^{-1}, 3256–3102 cm^{-1} and 3099–3037 cm^{-1}) and two bands for carbonyl function of ureido group were observed (1716–1670 cm^{-1} and 1545–1517 cm^{-1}). Apart from these a band for carbonyl of pyridazinone (1681–1654 cm^{-1}), a band for C=N of pyridazinone (1607–1589 cm^{-1}) and two bands for $SO_2N<$ (1350–1331 cm^{-1} and 1172–1136 cm^{-1}) were also observed. The structures were further established by proton NMR spectral data. A singlet for R-NH was observed at δ 5.68–6.56. Two doublets at δ 7.16–7.65 and δ 8.13–8.20 (J = 9.5–9.8 Hz) for the pyridazinone ring protons were also observed.

In IR spectra of **2a**–**e** bands at 3310–3042 cm^{-1} (NH), 1753–1736 cm^{-1} (C=O of carbamate), 1668–1656 cm^{-1} (cyclic carbonyl), 1356–1349 cm^{-1} and 1167–1160 cm^{-1} (SO_2N), 1240–1222 cm^{-1} and 1094–1089 cm^{-1} (C–O of carbamate) were identified. The ^{1}H NMR spectral data are in accordance with the structure of carbamates. A triplet at δ 1.06–1.13 and a quartet at δ 4.04–4.37 can be assigned to –OCH_2CH_3. Pyridazinone ring protons were observed at δ 7.23–7.29 and δ 8.17–8.23 (J = 9.6–9.8 Hz). Only two out of the five carbamates exhibited a broad singlet for NH (exchangeable proton) at δ 12.18 (**2c**) and at δ 12.27 (**2d**).

2.2. Blood glucose lowering effect

In the present study streptozotocin injection in neonatal rats induces in adult rats defects similar to those observed in mild type-II human diabetes (Non-insulin dependent diabetes mellitus) successfully [12]. NIDDM rats showed significantly higher glucose levels of fasting (>140 mg/dl) and glucose (2 g/kg) fed levels (>200 mg/dl two hours after oral administration of glucose). Oral antihyperglycemic efficacies of thirteen compounds (**3a**–**c**, **3e**, **3g**–**o**) were assessed using an oral glucose tolerance test in normal and NIDDM rat model. The marketed sulfonylurea drug gliclazide was selected as positive control. The sugar lowering effect of these compounds is presented in Table 1.

The compounds (**3a**–**c**, **3e**, **3g**–**o**) except **3c**, **3e** and **3o** at 20 mg/kg b.w (p.o.) almost completely prevented the rise of blood glucose of NIDDM rats as compared with NIDDM control. While compounds **3c** and **3o** showed more than 50% reduction in the rise of blood glucose levels. In glucose-fed normal rats these compounds at the same dose except **3e** significantly prevent the rise of blood glucose (more than 50%) when compared with control of glucose-fed normal rats.

The compound **3a** with phenyl group at C_6 of pyridazinone nucleus and propyl moiety in the side chain –NHR is the most active compound. The activity of **3a** became less when the bulk of 'R' was increased (**3a**–**c**). Introduction of Cl at *para* position of phenyl group caused slightly reduction in the activity (**3k** vs. **3a**). Activity of **3k** was further reduced when the bulk of 'R' was increased (**3k**–**n**). Compound **3f** with methoxyl moiety at *para* of phenyl group exhibited inferior activity. Presence of methyl moiety in phenyl group caused reduction in the activity (**3g** vs. **3a**, **3o** vs. **3m**). On the basis of these observations the SAR may be summarized as:

- In the side chain –NHR, less bulky 'R' seems more favourable for good activity.
- Presence of electron releasing moiety in phenyl group may cause the reduction in the activity.

It is well known that clinically used sulfonylureas exert antihyperglycemia action by stimulating insulin secretion by binding to and inhibiting the ATP-dependent K^+ channels in the ß-cell membrane, resulting ultimately in an opening of Ca^+ channels [13]. Since the synthesized derivatives are also sulfonylurea derivatives,

Table 1
Effect of sulfonylurea derivatives on blood glucose levels in normal and n2-STZ rats.

Compound	Ar/R	Blood glucose level (mg/dl) in normal rats		Percent inhibition in the rise of blood glucose level in normal rats	Blood glucose level (mg/dl) in n2-STZ rats		Percent inhibition in the rise of blood glucose level in n2-STZ rats
		Just prior to glucose loading (2 g/kg)	After 90 min of glucose loading (2 g/kg)		Just prior to glucose loading (2 g/kg)	After 90 min of glucose loading (2 g/kg)	
Control	–	146.3 ± 10.2	220.4 ± 5.6	–	77.3 ± 4.3	113.2 ± 5.2	–
Gliclazide	–	135.0 ± 9.1	109.0 ± 10.2	135	65.7 ± 3.2	63.3 ± 5.2	106.7
3a	C_6H_5/C_3H_7	143.2 ± 11.3	125.0 ± 11.6	124.6	73.5 ± 5.7	81.7 ± 5.7	77.2
3b	C_6H_5/C_4H_9	138.6 ± 14.2	130.2 ± 14.6	111.3	75.6 ± 4.3	87.3 ± 4.5	67.4
3c	C_6H_5/C_6H_{11}	140.3 ± 13.2	150.5 ± 17.2	86.2	70.0 ± 2.3	87.2 ± 5.4	52.1
3d	$C_6H_5/CH_2C_6H_5$	ND	ND	ND	ND	ND	ND
3e	4-$OCH_3C_6H_4/C_4H_9$	140.2 ± 10.2	195.3 ± 12.1	25.6	77.5 ± 5.5	97.7 ± 6.2	43.7
3f	4-$OCH_3C_6H_4/C_6H_{11}$	ND	ND	ND	ND	ND	ND
3g	4-$CH_3C_6H_4/C_3H_7$	142.2 ± 11.3	135.5 ± 12.2	109	75.3 ± 5.8	82.3 ± 6.3	80.5
3h	4-$CH_3C_6H_4/C_4H_9$	146.6 ± 14.3	140.0 ± 15.2	108.9	76.6 ± 5.7	81.4 ± 5.8	86.6
3i	4-$CH_3C_6H_4/C_6H_{11}$	137.7 ± 13.2	130.0 ± 10.2	110.4	77.4 ± 6.7	84.6 ± 6.1	79.9
3j	4-$CH_3C_6H_4/CH_2$-C_6H_5	139.3 ± 13.9	125.0 ± 12.2	119.3	76.2 ± 4.5	88.8 ± 5.2	64.9
3k	4-ClC_6H_4/C_3H_7	136.6 ± 13.6	120.5 ± 10.2	121.7	75.3 ± 5.8	84.5 ± 6.3	74.3
3l	4-ClC_6H_4/C_4H_9	140.4 ± 16.8	140.3 ± 11.7	100.1	77.7 ± 6.8	89.9 ± 7.2	66
3m	4-ClC_6H_4/C_6H_{11}	147.2 ± 9.7	136.6 ± 9.7	114.3	72.0 ± 3.3	86.7 ± 5.4	59
3n	4-ClC_6H_4/CH_2-C_6H_5	147.2 ± 10.2	139.0 ± 15.2	111.1	74.0 ± 4.3	87.8 ± 4.7	61.6
3o	4-Cl, 3-$CH_3C_6H_3/C_6H_{11}$	147.2 ± 10.2	180.4 ± 11.1	55.2	78.5 ± 6.5	92.4 ± 5.8	62.1

Values are mean ± SEM of 6 animals.
ND = not determined.

it may be assumed that the blood glucose lowering effect of these derivatives (**3a–c**, **3g–n**) is related to a stimulation of β-cells.

3. Conclusions

Our data suggested that eleven novel sulfonylurea derivatives (**3a–c**, **3g–n**) at the dose of 20 mg/kg exhibited considerably potent blood glucose lowering activity. These derivatives may be used as lead compounds for developing new antidiabetic drugs.

4. Experimental protocols

Melting points were determined by open capillary tubes and are uncorrected. All the Fourier Transform Infra Red (FTIR) spectra were recorded on Bio-rad FTS-135 spectrophotometer using KBr pellets; ν_{max} values are given in cm^{-1}. ^1H NMR and ^{13}C NMR spectra were recorded on Bruker Spectrospin DPX 300 MHz and Bruker Spectrospin DPX 75 MHz spectrometer respectively using DMSO as a solvent and trimethylsilane (TMS) as an internal standard. Chemical shifts are given in δ (ppm) scale and coupling constants (J values) are expressed in Hz. Mass spectra (MS) were scanned by affecting FAB ionization JEOL-JMS-DX 303 system, equipped with direct inlet probe system. The m/z values of the more intense peaks are mentioned. Purity of the compounds was checked on TLC plates (silica gel G), which were visualized by exposing to iodine vapours. Elemental analysis was carried out on CHNS Elementar (Vario EL III).

4.1. Chemistry

4.1.1. General procedure for the preparation of carbamates (*2a–e*)

A mixture of appropriate pyridazinone (**1a–e**) (0.01 mol), ethyl chloroformate (0.013 mol), and anhydrous potassium carbonate (2 g) in dry acetone (300–500 ml) was refluxed for 18 h. Acetone was removed by distillation under reduced pressure; the residue left was suspended in water (100 ml) and neutralized with acetic acid. The product separated out was filtered and washed with distilled water. The residue was dried and crystallized from methanol or acetone.

*4.1.1.1. Ethyl({4-[6-phenyl-3-oxopyridazin-2-yl]phenyl}sulfonyl)carbamate (**2a**).* Yield = 32.7%; m.p. 206–207 °C; IR ν_{max} (KBr, in cm^{-1}): 3042 (NH), 1753 (C=O of carbamate), 1664 (cyclic carbonyl), 1351 and 1167 (SO_2N), 1230 and 1089 (C–O of carbamate); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.06 (3H, t, CH_3), 4.11 (2H, q, OCH_2), 7.24 (1H, d, J = 9.7 Hz, H-4), 7.50–7.94 (9H, m, Ar–H), 8.18 (1H, d, J = 9.7 Hz, H-5); FAB-MS (m/z): 399 [M^+].

*4.1.1.2. Ethyl({4-[6-(4-methoxyphenyl)-3-oxopyridazin-2-yl]phenyl}sulfonyl)carbamate (**2b**).* Yield = 27.7%; m.p. 217–218 °C; IR ν_{max} (KBr, in cm^{-1}): 3310 (NH), 1747 (C=O of carbamate), 1656 (cyclic carbonyl), 1349 and 1161 (SO_2N), 1222 and 1094 (C–O of carbamate); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.06 (3H, t, CH_3), 3.82 (3H, s, OCH_3), 4.37 (2H, q, OCH_2), 7.04–8.16 (10H, m, Ar–H, H-4 and H-5); FAB-MS (m/z): 429 [M^+].

*4.1.1.3. Ethyl({4-[6-(4-methylphenyl)-3-oxopyridazin-2-yl]phenyl}sulfonyl)carbamate (**2c**).* Yield = 71.7%; m.p. 187–188 °C; IR ν_{max} (KBr, in cm^{-1}): 3305 (NH), 1736 (C=O of carbamate), 1656 (cyclic carbonyl), 1351 and 1166 (SO_2N), 1240 and 1092 (C–O of carbamate); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.13 (3H, t, CH_3), 2.37 (3H,s, $ArCH_3$), 4.05 (2H, q, OCH_2), 7.23 (1H, d, J = 9.7 Hz, H-4), 7.33 (2H, d, J = 7.2 Hz, Ar–H), 7.86 (2H, d, J = 7.5 Hz, Ar–H) 8.02 (4H, m, Ar–H), 8.17 (1H, d, J = 9.7 Hz, H-5), 12.18 (1H, brs, SO_2NH); FAB-MS (m/z): 413 [M^+].

*4.1.1.4. Ethyl({4-[6-(4-chlorophenyl)-3-oxopyridazin-2-yl]phenyl}sulfonyl)carbamate (**2d**).* Yield = 41.2%; m.p. 213–214 °C; IR ν_{max} (KBr, in cm^{-1}): 3065 (NH), 1753 (C=O of carbamate), 1668 (cyclic carbonyl), 1356 and 1167 (SO_2N), 1224 and 1092 (C–O of carbamate); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.12 (3H, t, CH_3), 4.06 (2H, q, OCH_2), 7.29 (1H, d, J = 9.7 Hz, H-4), 7.61 (2H, d, J = 8.2 Hz, Ar–H), 7.92–8.06 (6H, m, Ar–H), 8.23 (1H, d, J = 9.8 Hz, H-5), 12.27 (1H, brs, SO_2NH); FAB-MS (m/z): 434 [M + 1], 420.

*4.1.1.5. Ethyl({4-[6-(4-chloro-3-methylphenyl)-3-oxopyridazin-2-yl]phenyl}sulfonyl) carbamate (**2e**).* Yield = 26.6%; m.p. 203–204 °C; IR ν_{max} (KBr, in cm^{-1}): 3310 (NH), 1741 (C=O of carbamate), 1659 (cyclic carbonyl), 1350 and 1160 (SO_2N), 1231 and 1092 (C–O of carbamate); ^1H NMR (300 MHz, DMSO, δ): 1.13 (3H, t, CH_3), 2.40 (3H, s, $ArCH_3$), 4.04 (2H, q, OCH_2), 7.25 (1H, d, J = 9.6 Hz, H-4), 7.55 (1H, d, J = 8.2 Hz, Ar–H), 7.80 (1H, d, J = 7.8 Hz, Ar–H), 7.95–8.03 (5H, m, Ar–H), 8.18 (1H, d, J = 9.6 Hz, H-5); FAB-MS (m/z): 448 [M^+].

*4.1.2. General procedure for the preparation of pyridazinone substituted benzenesulfonylurea (**3a–o**)*

On complete dissolution of appropriate carbamates (**2a–o**) (0.001 mol) in boiling toluene (25–75 ml) was added the desired primary amine (0.0011 mol) dropwise. The mixture was subsequently refluxed for further 3–4 h and cooled. The product, which precipitated out on cooling, was filtered and crystallized from methanol.

*4.1.2.1. 1-[4-(6-Phenyl-3-oxopyridazin-2-yl)benzenesulfonyl]-3-(1-propyl)urea (**3a**).* M.p. 187–188 °C; yield 43.5%; IR ν_{max} (KBr, in cm^{-1}): 3390, 3192 and 3099 (NH of ureido group), 1716 and 1522 (C=O of urea), 1658 (cyclic carbonyl), 1596 (C=N), 1332 and 1162 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.78 (3H, t, CH_3), 1.36 (2H, m, $-CH_2-CH_2-CH_3$), 2.92 (2H, m, $HN-CH_2-CH_2-CH_3$), 6.54 (1H, brs, $NH-C_3H_7$), 7.24 (1H, d, $J = 9.8$ Hz, H-4), 7.50 (3H, m, Ar–H), 7.96 (4H, m, Ar–H), 8.04 (2H, d, $J = 8.5$ Hz, Ar–H), 8.18 (1H, d, $J = 9.8$ Hz, H-5), 10.61 (1H, br s, SO_2NH); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 11.12 (CH_3), 22.45 (CH_2), 40.96 (CH_2NH), 144.98 (C=N of pyridazinone), 151.34 (C=O), 158.62 (C=O); FAB-MS (m/z): 412 [M^+], 413 [M + 1].

*4.1.2.2. 1-[4-(6-Phenyl-3-oxopyridazin-2-yl)benzenesulfonyl]-3-(1-butyl)urea (**3b**).* M.p. 177–178 °C; yield 37.4%; IR ν_{max} (KBr, in cm^{-1}): 3320, 3256 and 3037 (NH of ureido group), 1691 and 1535 (C=O of urea), 1669 (cyclic carbonyl), 1595 (C=N), 1337 and 1172 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.82 (3H, t, CH_3), 1.19 (2H, m, $CH_2-CH_2-CH_2-CH_3$) 1.31 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 2.96 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 6.51 (1H, br s, $NH-C_4H_9$), 7.23 (1H, d, $J = 9.7$ Hz, H-4), 7.51 (3H, m, Ar–H), 7.96 (4H, m, Ar–H), 8.04 (2H, d, $J = 8.5$ Hz, Ar–H), 8.17 (1H, d, $J = 9.8$ Hz, H-5), 10.67 (1H, brs, SO_2NH); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 13.57 (CH_3), 19.34 (CH_3CH_2), 31.26 (CH_2CH_2NH), 144.95 (C=N of pyridazinone), 151.33 (C=O), 158.60 (C=O); FAB-MS (m/z): 426 [M^+], 427 [M + 1].

*4.1.2.3. 1-[4-(6-Phenyl-3-oxopyridazin-2-yl)benzenesulfonyl]-3-(cyclohexyl)urea (**3c**).* M.p. 261–262 °C; yield = 15.5%; IR ν_{max} (KBr, in cm^{-1}): 3312 and 3163 (NH of ureido group), 1656 (cyclic carbonyl), 1542 (C=O of urea), 1340 and 1166 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.01–1.88 (10H, m, cyclohexyl protons), 5.68 (1H, brs, $NH-C_6H_{11}$), 7.18–8.20 (11H, m, Ar–H, H-4 and H-5); FAB-MS (m/z): 452 [M^+], 453 [M + 1].

*4.1.2.4. 1-[4-(6-Phenyl-3-oxopyridazin-2-yl)benzenesulfonyl]-3-(benzyl)urea (**3d**).* M.p. 201–202 °C; yield = 19.5%; IR ν_{max} (KBr, in cm^{-1}): 3330 (NH of ureido group), 1699 and 1523 (C=O of urea), 1655 (cyclic carbonyl), 1589 (C=N), 1335 and 1164 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 4.16 (2H, d, $J = 4.9$ Hz, $CH_2-C_6H_5$), 6.97–8.02 (15H, m, Ar–H and H-4), 8.19 (1H, d, $J = 9.8$ Hz, H-5); FAB-MS (m/z): 460 [M^+], 461 [M + 1].

*4.1.2.5. 1-[4-{6-(4-Methoxyphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(1-butyl)urea (**3e**).* M.p. 167–168 °C; yield = 41.1%; IR ν_{max} (KBr, in cm^{-1}): 3313, 3243 and 3062 (NH of ureido group), 1677 and 1518 (C=O of urea), 1654 (cyclic carbonyl), 1607 (C=N), 1340 and 1170 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.83 (3H, t, CH_3), 1.20 (2H, m, $CH_2-CH_2-CH_2-CH_3$) 1.31 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 2.95 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 3.82 (3H, s, OCH_3), 6.54 (1H, brs, $NH-C_6H_{11}$), 7.05 (2H, d, $J = 8.1$ Hz, Ar–H), 7.21 (1H, d, $J = 9.7$ Hz, H-4), 7.90–8.04 (6H, m, Ar–H), 8.16 (1H, d, $J = 9.8$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 13.58 (CH_3), 19.36 (CH_3CH_2), 31.28 (CH_2CH_2NH), 55.36 (OCH_3) 145.03 (C=N of pyridazinone), 151.40 (C=O), 158.51 (C=O), 160.62 (Ar C–O–C); FAB-MS (m/z): 456 [M^+].

*4.1.2.6. 1-[4-{6-(4-Methoxyphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(cyclohexyl)urea (**3f**).* M.p. 269–270 °C; yield = 15%; IR ν_{max} (KBr, in cm^{-1}): 3097 (NH of ureido group), 1517 (C=O of urea), 1681 (cyclic carbonyl), 1589 (C=N), 1333 and 1136 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.00–1.87 (10H, m, cyclohexyl protons), 3.81 (3H, s, OCH_3), 5.70 (1H, br s, $NH-C_6H_{11}$), 7.04 (2H, d, $J = 7.9$ Hz, Ar–H), 7.16 (1H, d, $J = 9.6$ Hz, H-4), 7.62–7.91 (6H, m, Ar–H), 8.13 (1H, d, $J = 9.5$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 24.81 ($2 \times CH_2$), 25.40 (CH_2), 33.34 ($2 \times CH_2$), 49.35 (CH–NH), 144.10 (C=N of pyridazinone), 158.47 (C=O), 159.36 (C=O), 160.46 (Ar C–O–C); FAB-MS (m/z): 482 [M^+], 483 [M + 1], 460.

*4.1.2.7. 1-[4-{6-(4-Methylphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(1-propyl)urea (**3g**).* M.p. 167–168 °C; yield 48%; IR ν_{max} (KBr, in cm^{-1}): 3351, 3102 (NH of ureido group), 1670 (cyclic carbonyl), 1544 (C=O of urea), 1604 (C=N), 1350 and 1168 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.77 (3H, t, CH_3), 1.35 (2H, m, $-CH_2-CH_2-CH_3$), 2.36 (3H, s, $C_6H_5CH_3$), 2.92 (2H, m, $HN-CH_2-CH_2-CH_3$), 6.55 (1H, t, $NH-C_3H_7$), 7.22 (1H, d, $J = 9.8$ Hz, H-4), 7.32 (2H, d, $J = 7.9$ Hz, Ar–H), 7.85 (2H, d, $J = 8.0$ Hz, Ar–H), 7.95 (2H, d, $J = 8.6$ Hz, Ar–H), 8.04 (2H, d, $J = 8.6$ Hz, Ar–H), 8.16 (1H, d, $J = 9.8$ Hz, H-5), 10.70 (1H, br s, SO_2NH); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 11.13 (CH_3), 20.89 ($ArCH_3$), 22.47 (CH_2), 41.0 (CH_2NH), 145.04 (C=N of pyridazinone), 151.4 (C=O), 158.64 (C=O); FAB-MS (m/z): 426 [M^+], 427 [M + 1], 368, 342 and 278.

*4.1.2.8. 1-[4-{6-(4-Methylphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(1-butyl)urea (**3h**).* M.p. 201–202 °C; yield 47%; IR ν_{max} (KBr, in cm^{-1}): 3305 and 3217 (NH of ureido group), 1697 and 1536 (C=O of urea), 1675 (cyclic carbonyl), 1604 (C=N), 1334 and 1162 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.81 (3H, t, CH_3), 1.19 (2H, m, $CH_2-CH_2-CH_2-CH_3$) 1.31 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 2.37 (3H, s, $C_6H_5CH_3$), 2.96 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 6.52 (1H, br s, $NH-C_4H_9$), 7.22 (1H, d, $J = 9.8$ Hz, H-4), 7.32 (2H, d, $J = 7.9$ Hz, Ar–H), 7.85 (2H, d, $J = 8.0$ Hz, Ar–H), 7.95 (2H, d, $J = 8.6$ Hz, Ar–H), 8.04 (2H, d, $J = 8.6$ Hz, Ar–H), 8.15 (1H, d, $J = 9.8$ Hz, H-5), 10.30 (1H, br s, SO_2NH); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 13.61 (CH_3), 19.41 (CH_3CH_2), 20.90 ($ArCH_3$), 31.33 (CH_2CH_2NH), 145.07 (C=N of pyridazinone), 151.44 (C=O), 158.66 (C=O); FAB-MS (m/z): 440 [M^+], 441 [M + 1], 368, 342 and 278.

*4.1.2.9. 1-[4-{6-(4-Methylphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(cyclohexyl)urea (**3i**).* M.p. 189–190 °C; yield = 55.7%; IR ν_{max} (KBr, in cm^{-1}): 3367, 3250 and 3080 (NH of ureido group), 1675 (cyclic carbonyl), 1520 (C=O of urea), 1602 (C=N), 1338 and 1159 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.09–1.88 (10H, m, cyclohexyl protons), 2.35 (3H, s, $C_6H_5CH_3$), 6.13 (1H, brs, $NH-C_6H_{11}$), 7.30 (2H, m, Ar–H), 7.65 (1H, d, $J = 9.6$ Hz, H-4), 7.83–7.94 (6H, m, Ar–H), 8.14 (1H, d, $J = 9.6$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 20.90 ($ArCH_3$), 24.58 ($2 \times CH_2$), 25.24 (CH_2), 32.83 ($2 \times CH_2$), 49.44 (CH–NH), 144.61 (C=N of pyridazinone), 154.95 (C=O), 158.66 (C=O); FAB-MS (m/z): 466 [M^+], 467 [M + 1], 385, 368, 342 and 278.

*4.1.2.10. 1-[4-{6-(4-Methylphenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(benzyl)urea (**3j**).* M.p. 209–210 °C; yield = 34%; IR ν_{max} (KBr, in cm^{-1}): 3345, 3247 and 3066 (NH of ureido group), 1701 and 1527 (C=O of urea), 1676 (cyclic carbonyl), 1602 (C=N), 1333 and 1162 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 2.37 (3H, s, $C_6H_5CH_3$), 4.18 (2H, s, $CH_2-C_6H_5$), 7.07–8.19 (15H, m, Ar–H, H-4 and H-5), 10.92 (1H, br s, SO_2NH); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 20.85 ($ArCH_3$), 42.78 ($ArCH_2$), 144.98 (C=N of pyridazinone), 151.76 (C=O), 158.56 (C=O); FAB-MS (m/z): 474 [M^+], 475 [M + 1], 441, 342 and 262.

*4.1.2.11. 1-[4-{6-(4-Chlorophenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(1-propyl)urea (**3k**).* M.p. 177–178 °C; yield 30%; IR ν_{max}

(KBr, in cm^{-1}): 3310, 3236 and 3041 (NH of ureido group), 1698 and 1534 (C=O of urea), 1673 (cyclic carbonyl), 1600 (C=N), 1335 and 1168 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.77 (3H, t, CH_3), 1.35 (2H, m, $-CH_2-CH_2-CH_3$), 2.91 (2H, m, $HN-CH_2-CH_2-CH_3$), 6.56 (1H, t, $NH-C_3H_7$), 7.25 (1H, d, $J = 9.8$ Hz, H-4), 7.58 (2H, d, $J = 8.5$ Hz, Ar–H), 7.94–8.05 (6H, m, Ar–H), 8.19 (1H, d, $J = 9.8$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 11.15 (CH_3), 22.49 (CH_2), 41.02 (CH_2NH), 144.92 (C=N of pyridazinone), 151.46 (C=O), 158.61 (C=O); FAB-MS (m/z): 447 [M^+], 391 and 362.

*4.1.2.12. 1-[4-{6-(4-Chlorophenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(1-butyl)urea (**3l**).* M.p. 207–208 °C; yield 38%; IR ν_{max} (KBr, in cm^{-1}): 3313, 3236 and 3063 (NH of ureido group), 1670 and 1542 (C=O of urea), 1673 (cyclic carbonyl), 1602 (C=N), 1337 and 1164 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 0.82 (3H, t, CH_3), 1.20 (2H, m, $CH_2-CH_2-CH_2-CH_3$) 1.31 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 2.93 (2H, m, $HN-CH_2-CH_2-CH_2-CH_3$), 6.43 (1H, br s, $NH-C_4H_9$), 7.24 (1H, d, $J = 9.8$ Hz, H-4), 7.56 (2H, d, $J = 8.4$ Hz, Ar–H), 7.90 (2H, d, $J = 8.4$ Hz, Ar–H), 7.99 (4H, m, Ar–H), 8.18 (1H, d, $J = 9.8$ Hz, H-5). ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 13.64 (CH_3), 19.42 (CH_3CH_2), 31.34 (CH_2CH_2NH), 144.93 (C=N of pyridazinone), 151.49 (C=O), 158.64 (C=O); FAB-MS (m/z): 461 [M^+], 388 and 362.

*4.1.2.13. 1-[4-{6-(4-Chlorophenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(cyclohexyl)urea (**3m**).* M.p. 281–282 °C; yield = 38%; IR ν_{max} (KBr, in cm^{-1}): 3308, 3248 and 3066 (NH of ureido group), 1703 and 1544 (C=O of urea), 1681 (cyclic carbonyl), 1601 (C=N), 1344 and 1168 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 1.10–1.85 (10H, m, cyclohexyl protons), 6.39 (1H, br s, $NH-C_6H_{11}$), 7.23 (1H, d, $J = 9.8$ Hz, H-4), 7.55 (2H, d, $J = 8.5$ Hz, Ar–H), 7.90–8.02 (6H, m, Ar–H), 8.16 (1H, d, $J = 9.8$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 24.61 ($2 \times CH_2$), 25.29 (CH_2), 33.0 ($2 \times CH_2$), 49.40 (CH–NH), 143.95 (C=N of pyridazinone), 156.5 (C=O), 158.59 (C=O); FAB-MS (m/z): 487 [M^+], 405, 391, 362.

*4.1.2.14. 1-[4-{6-(4-Chlorophenyl)-3-oxopyridazin-2-yl}benzenesulfonyl]-3-(benzyl)urea (**3n**).* M.p. 215–216 °C; yield = 24%; IR ν_{max} (KBr, in cm^{-1}): 3346, 3250 and 3066 (NH of ureido group), 1702 and 1528 (C=O of urea), 1678 (cyclic carbonyl), 1600 (C=N), 1331 and 1162 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): 4.18 (2H, d, $CH_2-C_6H_5$), 7.06–7.28 (6H, m, Ar–H and H-4), 7.58 (2H, d, $J = 8.5$ Hz, Ar–H), 7.96–8.06 (6H, m, Ar–H), 8.19 (1H, d, $J = 9.8$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 42.87 ($ArCH_2$), 144.90 (C=N of pyridazinone), 152.0 (C=O), 158.65 (C=O); FAB-MS (m/z): 495 [M^+], 452, 387, 362, 281(base peak).

*4.1.2.15. 1-[4-{6-(4-Chloro-3-methylphenyl)-3-oxopyridazin-2-yl} benzenesulfonyl]-3-(cyclohexyl)urea (**3o**).* M.p. 274–275 °C; yield = 15.1%; IR ν_{max} (KBr, in cm^{-1}): 3379 (NH of ureido group), 1707 and 1545 (C=O of urea), 1663 (cyclic carbonyl), 1604 (C=N), 1339 and 1160 (SO_2N); ^1H NMR (300 MHz, DMSO-d_6, δ): δ: 1.13–1.65 (10H, m, cyclohexyl protons), 2.39 (3H, s, CH_3), 6.38 (1H, br s, $NH-C_6H_{11}$), 7.24 (1H, d, $J = 9.8$ Hz, H-4), 7.49 (1H, d, $J = 8.4$ Hz, Ar–H), 7.80 (1H, d, $J = 7.9$ Hz, Ar–H), 7.91–8.04 (5H, m, Ar–H), 8.20 (1H, d, $J = 9.7$ Hz, H-5); ^{13}C NMR (75 MHz, DMSO-d_6, δ, ppm): 19.65 ($ArCH_3$), 24.24 ($2 \times CH_2$), 24.98 (CH_2), 32.30 ($2 \times CH_2$), 48.20 (CH–NH), 144.86 (C=N of pyridazinone), 150.55 (C=O), 158.55 (C=O); FAB-MS (m/z): 501 [M^+].

4.2. Blood glucose lowering studies

In the present study effects of oral administration of synthesized sulfonylurea derivatives on glucose tolerance in normal and streptozotocin (STZ) induced NIDDM rats have been investigated.

4.2.1. Induction of non-insulin dependent diabetes mellitus in rats

Healthy albino female wistar rats were kept for breeding. To induce NIDDM, streptozotocin (STZ) (100 mg/kg) freshly dissolved in citrate buffer solution (0.1 M, pH 4.5) was administered intraperitoneally to 2-day old pups weighing 6–8 g. The pups were weaned for 4 weeks and separated thereafter from their mothers. Male and female rats were separately housed with food and water *ad libitum*. When the rats were 12 weeks old the animals were checked for fasting glucose levels. The animals showing fasting glucose levels >140 mg/dl and 200 mg/dl 2 h after 2 g/kg oral challenge of glucose were considered as diabetic.

4.2.2. Effect of synthesized compounds on oral glucose tolerance in diabetic (n2-STZ) rats

Albino rats (either sex) of wistar strain weighing 160–200 g were fasted overnight and classified into groups of six animals each. Group I served as control received vehicle (1% CMC in distilled water) in a volume of 10 ml/kg. The reference drug gliclazide (20 mg/kg) and synthesized compounds (**3a–c**, **3e**, **3g–o**) in the dose of 20 mg/kg suspended in vehicle were administered p.o. in a volume of 10 ml/kg to respective groups. All the animals were given glucose (2 g/kg p.o.) 15 min after dosing. Blood samples were collected from the retro-orbital plexus just prior to and 90 min after the glucose loading and glucose levels were measured (Table 1).

4.2.3. Effect of synthesized compounds on oral glucose tolerance in normal rats

The same protocol as mentioned in above experiment was adopted for this experiment. Instead of n2-STZ rats, normal rats were used (Table 1).

4.2.4. Blood glucose estimation

The blood glucose level was assayed by an enzymatic colorimetric method using a commercially available glucose oxidase–peroxidase (GOD–POD) kit purchased from Span diagnostics Ltd., Surat, India. Glucose oxidase (GOD) oxidizes glucose to gluconic acid and hydrogen peroxide. In presence of enzyme peroxidase, released hydrogen peroxide is coupled with phenol and 4-amino-antipyrine (4-AAP) to form coloured quinoneimine dye. Absorbance of coloured dye is measured at 505 nm and is directly proportional to glucose concentration in the sample. It adheres to the Beer–Lambert law over a wide range of glucose concentration.

4.2.5. Calculation

Results of blood glucose estimation are presented in the form of ±SEM and percent reduction by test drug. The percent reduction was calculated by considering the difference between the blood glucose levels of respective control animals at just prior to glucose loading and at 90 min after of glucose loading as 100% rise in the blood sugar. A 100% reduction indicates there is no rise in the level of blood sugar, while 0% reduction indicates there is no reduction in the level of blood sugar.

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