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P Balakrishnaiah
Vignan Institute of Pharmaceutical Technology, Duvvada, Visakhapanam, A.P, India

T Satyanarayana
College of Pharmaceutical Sciences, Andhra University, Visakhapanam, A.P, India

Correspondence
P Balakrishnaiah
Vignan Institute of Pharmaceutical Technology, Duvvada, Visakhapanam, A.P, India

Comparative study of antidiabetic activity among three medicinal plants against streptozotocin induced diabetes rat model

P Balakrishnaiah and T Satyanarayana

Abstract
Three plants from indian origin like aerial parts of *Schrebera swietenoides,* roots of *Barleria montana* and aerial parts of *Rotula aquatica* were extracted with methanol and metanolic extracts were evaluated for antidiabetic activity against streptozotocin induced diabetes for their study. Oral administration of these plant extracts at different dose levels of 100 mg/kg, 200 mg/kg and 400 mg/kg were screened in comparision with the standard drug glibenclimide. Among these plant extracts, extract of *Barleria montana* at a dose of 400 mg/kg b.w exhibited significant activity within 4th and 8th hour intervals showing a reduction in blood glucose levels are 293.94± 4.63 and 235.04± 2.93 mg/dl. preliminary phytochemical screening also conducted which revealed presence of triterpenes, flavonoids and steroid compounds.

Keywords: *Barleria montana, Rotula aquatica, Schrebera swietenoides,* antidiabetic activity

1. Introduction
Endocrine portion of pancreas participate a crucial role in the metabolism of active molecules of carbohydrates, lipids and proteins for their function in the body. Improper metabolism of these substances with the insufficient release of insulin led to the formation of diabetes characterized by hyperglycemia and hypoinsulimia. Even with the continuous availability of synthetic drugs in the market sources from the natural drugs has been paid much attention as these possess good activity with no side effects than the former one [1, 2].
Schrebera swietenoids is distributed in the hills of dry deciduous forests at 600-1000 m. The root of the plant is used for the treatment of leprosy and diabetes. Application of root paste on throat and chest produces relief for the obstruction of nasal cavity within the respiratory tract [3,4]. *Barleria montana* (F: Acanthaceae) plant is amply known for its correspondence of Barleria purprea which is completely developed on hills of helpless slopes, plains amid rocks and at higher elevations. Leaf of the concoct isrecognized from the harsh times for its review in the benefit of diabetes, wounds, and cuts [5,6]. Pashanabhedah is the common cast for Rotula aquatica is inasmuch as of expressing its chance in dissolving stones from kidney. It has been smoothly recommended in the service of cough, cardiac disorder, blood disorders, fever and ulcers [7].
Due to lack of establishment in proper antidiabetic activity upon the plants and increase in demand for outcome of effective drugs from the natural sources within the formulation, the selected plants were evaluated for activity against streptozotocin induced diabetic rats.

2. Materials and Methods
The engaged plants were extracted by the whole of methanol and the resultant extracts were suspended in 1% Sodium CMC and administered at a dose levels of 100, 200 and 400 mg/kg. The author hand me down STZ obsessed diabetes as a epitome to consider the antidiabetic transpire of all by one lonesome extracts. In the present study the Wistar Albino rats of either sex supplied by Mahaveer Enterprises, Hyderabad, weighing 150 to 200 g were used.
Aerial parts of schrebera swietenodes, roots of Barleria montana and aerial parts of Rotula aquatica were collected from various parts of India during winter season. Crude forms of the drugs were grounded in wilay mill for subjection toward extraction with a soxhlet apparatus as solvent methanol. The plants were authenticated with the help of a botanist Dr Venklaiah, Andhra University, Visakhapatnam and the voucher specimens from Aerial parts of schrebera swietenodes (AU/SS/TSN/IND/029), roots of Barleria montana(AU/ BM/TSN/IND/030) and aerial parts of Rotula aquatic (AU/RA/TSN/IND/031) were procured in the department of Pharmacognosy and Phytochemistry, University college of Pharmaceutical Sciences, Andhra.

University, Visakhapatnam. The resultant extracts after the extraction were subjected to various chemical tests for presence of compounds.

2.1. Acute toxicity studies

Toxicity studies were conducted as by the agency of internationally implied protocol dead on one feet under OECD guidelines in Wistar albino rats at a dose in a new york minute of extracts like a one man band 2000 mg/kg b.w. The toxic chance of the methanolic extracts of *Schrebera swietenoides* (aerial parts), *Barleria montana* (roots) and *Rotula aquatica* (aerial parts) were clocked in punched in at a dose easily of 2000 mg/kg b.w. The animals were further closely given right consideration for signs of intoxication, torpor, behavioral transformation and morbidity [8].

2.2. Screening of extracts for Antihyperglycemic activity

Methanolic extracts of selected plants were camouflaged for antihyperglycemic big idea in STZ obsessed diabetic rats at doses of 100, 200 and 400 mg/kg b.w. The drug benefit was subject to to the animals and was fasted for 12 hr already estimating the ties of flesh brother blood level. The animals were allowed to secure food and raw material ad libitum. The same order of the day of management was followed for booze treated accumulation (vehicle control) and Glibenclamide (0.45 mg/kg b.w). The ties of blood brother samples were united from the retro orbital plexus of rats. After group of blood tried the flavor of, the serum was living alone by centrifuge at 3000 rpm for 10 mins as cleanly as serum glucose levels were estimated at 0, 2, 4, 8, 12, 18, and 24th hr. The percent blood glucose loss of value was besides calculated individually group.

2.3. Experimental design

In this demonstrate, 66 rats were hand me down which were randomly isolated into 11 groups of 6 animals each. The disparate doses of extracts were administered oral message to the STZ possessed diabetic rats. All the extracts were suspended in 1% sodium CMC suspension. In these 11 groups, one everything served as untreated approach as they confirmed orally 1% Sodium CMC suspension unattended and one accumulation received standard abused substance Glibenclamide (0.45mg /kg b.w).

Group 1: Vehicle control (1% Sodium CMC suspension)
Group 2: Received extract of *Schrebera swietenoides* (100 mg/kg)
Group 3: Received extract of *Schrebera swietenoides* (200mg/kg)
Group 4: Received extract of *Schrebera swietenoides* (400mg/kg)
Group 5: Received extract of *Barleria montana* (100mg/kg)
Group 6: Received extract of *Barleria montana* (200mg/kg)
Group 7: Received extract of *Barleria montana* (400mg/kg)
Group 8: Received extract of *Rotula aquatica* (100 mg/kg)
Group 9: Received extract of *Rotula aquatica* (200 mg/kg)
Group 10: Received extract of *Rotula aquatica* (400 mg/kg)
Group 11: Standard (Glibenclamide 0.45mg/kg)

2.4. Induction of Diabetes

Diabetes was obsessed by a base hit intraperitonial dose of 60 mg/kg of b. w of streptozotocin (STZ) dissolved in 0.1M fresh deadpan citrate level of economic security guaranteed by government (pH 4.5) directed toward 12 hr fasted rats. On third many a moon of the STZ hypodermic of the rats, the ties of family brother samples were taken from retro orbital plexus of the rats for the approximation of blood blood levels by by the agency of the bus analyzer. Rats by all of diabetes having hyperglycemia (i.e. by all of blood glucose of 185 to 460 mg/dl) were taken for the experiment. The diabetic rats were before randomly sovereign into march to a different drummer groups [9-12].

2.5. Collection of blood samples and serum glucose estimation

The basal flesh glucose directly was enthusiastic by obtaining 0.5ml of ties of ties of blood brother brother from creep tail dissuade by by one art an adjunct of ultra glucometer (Life Scan Inc, Milpital, USA). The one apply strip was in into a glucometer and the tryout was noted [13]. The blood samples (0.5ml) were brought together for every anticipate intervels of 0, 2, 4, 8, 12, 18, and 24th hr in 1ml Eppendorf's tubes. Serum was unmarried by centrifuging at 3000 rpm for 10 minutes. 30 µl of serum tried the flavor of and 3 ml of occupied glucose reagent were simple to a restrained and above suspicion test tv set and incubated for 10 minutes at 37o C. The pink blew up out of proportion developed was measured by per auto analyzer [14-16]

2.6. Statistical Analysis

The values were expressed as mean ±SEM. The data was subjected to the analysis of variance (one way ANOVA) to determine the significance of changes followed by students "t"-test. The statistical significance of difference between two independent groups was calculated for the determination of blood glucose levels [17].

3. Results & Discussion

The mean blood glucose levels of control and drug treated animals after oral administration of different doses (100, 200 and 400 mg/kg b.w) of methanolic extracts of *Schrebera swietenoides* aerial parts, *Barleria montana* roots and aerial parts of *Rotula aquatica* at various time intervals (0, 2, 4, 8, 12, 18 and 24 hrs) were shown in Table 1 and Figure 1. The statistical significance of decrease in blood glucose levels was calculated with respect to initial blood glucose levels. Oral administration of 1% Sodium CMC suspension did not change the blood glucose levels of rats.

The blood glucose levels of diabetic rats treated with Glibenclamide (0.45 mg/kg b.w) showed significant ($P<0.05$) decrease in blood glucose levels at 8 &12th hrs, more significant ($P<0.01$) decrease in blood glucose levels at 2nd hr and highly significant ($P<0.001$) decrease in blood glucose levels at 4th hr. Nevertheless, the reduction in mean blood glucose levels was no significant at 18 & 24 hrs. After the oral administration of standard drug the mean blood glucose levels were 353.29±12.13,261.20±8.52, 201.93±5.24, 274.68±15.41 and 302.52±3.47 mg/dl at 0, 2, 4, 8 and 12th hr respectively.

Administration of 100 and 200 mg/kg b. w of plant crude drug produced no significant (P>0.05) decrease in blood glucose levels at all the time intervals. The oral administration of 400 mg/kg b. w of methanolic extract of *Schrebera swietenoides* aerial parts showed highly significant ($P<0.001$) decrease in blood glucose levels at 8th hr respectively. The mean blood glucose levels were 348.14± 6.11, 290.61±9.80, 220.56± 9.61, 283.43± 9.11 and 297.52± 11.65 mg/dl at 0, 4, 8, 12 and 18th hr respectively after the administration of 400 mg/kg b. w of

methanolic extract of *Schrebera swietenoides* aerial parts. Oral administration of 400 mg /kg b. w of methanolic extract of *Barleria montana* roots showed significant ($P<0.05$) decrease in blood glucose levels at 2 & 12th hr and highly significant ($P<0.001$) decrease in blood glucose levels at 4th and 8th hr. The mean blood glucose levels 0, 2, 4, 8 and 12 hrs. After oral administration of 400 mg/kg b. w of methanolic extract of *Barleria montana* roots were 359.02±6.17, 319.00±9.75, 293.94±4.63, 235.04±293 and 304.27±18.89 mg/dl respectively. The oral administration of 400 mg/kg b.w of methanolic extract of aerial parts of *Rotula aquatica* showed significant ($P<0.05$) decrease in blood glucose levels at 2nd and 18th hrs, more significant ($P<0.01$) decrease in blood glucose levels at 4 and 8th hrs and highly significant ($P<0.001$) decrease in blood glucose levels at 8th hr. The mean blood glucose levels after oral administration of 400 mg/kg b. w of methanolic extract of aerial parts of *Rotula aquatica* were 355.29±8.09, 328.15±6.65, 295.04±9.15, 261.13±6.97, 291.59±5.50 and 318.50±5.13 mg/dl at 0, 2, 4, 8, 12 and 18th hrs respectively.

The percent decrease in blood glucose levels after the oral administration of different doses (100, 200 and 400 mg/kg b.w) of methanolic extracts of *Schrebera swietenoides* aerial parts, *Barleria montana* roots and aerial parts of *Rotula aquatic* was shown in Table 2 and Fig. 2. The administration of standard drug Glibenclamide showed 25.47±4.23 %, 42.41±2.94 %, 21.65±5.62 % and 13.81±3.41 % reduction in blood glucose levels at 2, 4, 8 and 12 hrs respectively. The percent decrease in blood glucose level at 24th hr after the administration of Glibenclamide was not significant ($P>0.05$).

Administration of 400 mg/kg b. w of methanolic extract of *Schrebera swietenoides* aerial parts produced significant ($P<0.05$) decrease in blood glucose level at 18 and 24th hr, more significant ($P<0.01$) decrease in blood glucose levels at 4 and 12th hr and highly significant ($P<0.001$) decrease in blood glucose levels at 8th hr. Administration of 400 mg/kg b. w of methanolic extract of *Schrebera swietenoides* aerial parts showed 16.39±3.28%, 36.46±3.35%, 18.46±2.95%, 14.37±3.82% and 10.13±3.68% reduction in blood glucose levels at 4, 8, 12, 18 and 24 hrs respectively. The maximum percent reduction in blood glucose levels was observed at 12th and 8th hr after oral administration of the 200 and 400 mg/kg b. w of methanolic extract of *Schrebera swietenoides* aerial parts.

The oral administration of 400 mg/kg b. w of methanolic extract of *Barleria montana* roots produced significant ($P<0.05$) decrease in blood glucose level at 2, 12, and 18 hr and highly significant ($P<0.01$) decrease in blood glucose level at 4 and 8th hrs. The mean percent decrease in blood glucose levels produced by 400 mg/kg b. w of methanolic extract of *Barleria montana* roots were 10.96±3.51%, 18.01±1.87%, 34.45±1.31%, 15.33±4.75% and 9.75±3.80% at 2, 4, 8, 12 and 18 hrs respectively. Oral management of 400 mg/kg b. w of methanolic extract of aerial parts of *Rotula aquatica* produced highly significant ($P<0.001$) percent reduction in blood glucose levels at 4,8,12 and 18th hrs compared to the control group at identical times. The mean percent decrease in blood glucose levels produced by 400 mg/kg b. w of methanolic extract of aerial parts of *Rotula aquatica* were 8.40±2.94%, 17.86±1.78%, 27.26±1.58%, 18.74±2.16% and 11.26±0.85% at 2, 4, 8, 12, and 18th hr respectively.

Streptozotocin (STZ) is as a matter of course used for experimental computer aided instruction of type-I diabetes mellitus, which details selective pancreatic islet β-cell cytotoxicity mediated over the preserve of nitric oxide (NO).This results in brisk reduction in pancreatic islet pyridine nucleotide deep thought and imminent β-cell necrosis. The develop of STZ on mitochondria generates SOD anions, which accelerate diabetic complications [18]. In our diamond in the rough, consistent decreased in flesh glucose and pick up in plasma insulin levels were observed in diabetic rats treated mutually *Barleria montana* methanol dig in to the past than the disparate two extracts. This could be merit to the applied force by the get of the pancreatic secretion of insulin constitute regenerated β-cells or its materialize to release rush insulin from regenerated β-cells by inhibiting ATP unofficial K+ channel relish Glibenclamide. Previous raw material showed that phenolic compounds acted on ATP for no other ears K+ channels and tidy blood glucose [19].

Table 1: Effect of Methanol extracts of selected plants on blood glucose levels (mg/dl) in STZ induced diabetic rats at doses of 100,200 and 400 mg/kg b.w.

Group (n=6)	Treatment mg/kg b.w.	Time in hours						
		0	2	4	8	12	18	24
1	Control	352.26± 14.13	343.53± 12.32	343.00± 14.68	341.09± 12.03	335.06± 11.37	341.05±11.95	339.63±10.76
2	Glibenclamide (0.45 mg/kg b.w.)	353.29 ±12.13	261.20± 8.52**	201.93± 5.24***	274.68± 15.41*	302.52 ±3.47*	323.76± 5.00	333.85± 7.09
3	SS 100	342.89± 7.45	327.53± 5.59	320.84 ± 5.28	318.99± 7.90	320.70± 6.89	318.82± 6.4	318.42± 8.18
4	SS 200	357.34± 15.22	344.34± 13.50	317.91± 7.58	289.13± 7.62*	281.68 ±9.34**	292.41± 5.94*	318.75± 3.89
5	SS 400	348.14 ±6.11	322.34± 10.59	290.61± 9.80**	220.56± 9.61***	283.43± 9.11**	297.52± 11.65*	312.96± 14.23
6	BM 100	345.16± 11.23	321.53± 9.94	301.86 ± 11.57*	278.6± 7.84**	314.11± 6.30	322.88± 4.28	339.00± 2.94
7	BM 200	339.65± 8.93	310.85± 6.05*	294.8± 7.5*	288.30± 7.76**	263.95 ±3.3***	283.65± 16.94*	282.48± 19.59*
8	BM 400	359.02 ±6.17	319.00± 9.75*	293.94± 4.63***	235.04± 2.93***	304.27± 18.89*	324.19± 15.46	332.96± 12.42
9	RA 100	339.7± 12.4	333.87± 7.19	321.92 ± 5.58	302.37± 4.75*	304.94± 6.73	309.69± 4.37	313.70± 5.94
10	RA200	343.24± 11.42	333.51± 9.13	324.02± 4.46	301.79± 4.97*	282.16 ±7.44**	310.42± 5.68*	324.08± 10.64
11	RA400	355.29 ±8.09	328.15± 6.65*	295.04± 9.15**	261.13± 6.97***	291.59± 5.50**	318.50± 5.13*	344.54± 3.91

Table 2: Effect of Methanol extracts of selected medicinal plants on percent decrease blood glucose levels in STZ induced diabetic rats

Group (n=6)	Treatment mg/kg b.w.	Time in hours					
		2	4	8	12	18	24
1	Control	2.30±2.02	2.48±2.83	2.90±2.61	4.54±2.92	2.91±2.55	3.29±2.34
2	Glibenclamide (0.45 mg/kg b.w)	25.47±4.23**	42.41±2.94***	21.65±5.62*	13.81±3.41**	7.59±4.60	4.87±4.18
3	SS 100	4.37±1.60*	6.34±1.21**	6.8±2.8	6.15±3.34	6.69±3.34	6.93±3.04
4	SS 200	3.52±1.19*	10.62±2.16**	18.45±3.73**	20.5±4.18**	17.28±4.57*	9.74±5.05
5	SS 400	7.32±3.24	16.39±3.28**	36.46±3.35***	18.46±2.95**	14.37±3.82*	10.13±3.68*
6	BM 100	6.34±4.36	11.98±4.97	18.63±4.55**	8.34±4.27	5.87±3.76	1.18±3.88
7	BM 200	8.32±1.82**	13.13±1.63***	14.41±3.92*	22.03±2.19***	15.82±6.52	16.05±7.42
8	BM 400	10.96±3.51*	18.01±1.87***	34.45±1.31***	15.33±4.75*	9.75±3.80*	7.22±3.30
9	RA 100	2.73±3.79	6.13±4.15	11.90±3.44*	10.99±4.46	9.66±4.02	8.51±4.08
10	RA 200	1.63±2.78	4.32±2.56	10.85±2.73*	16.58±3.47**	8.30±2.91*	4.18±4.37
11	RA 400	8.40±2.94*	17.86±1.78***	27.26±1.58***	18.74±2.16***	11.26±0.85***	3.91±1.87

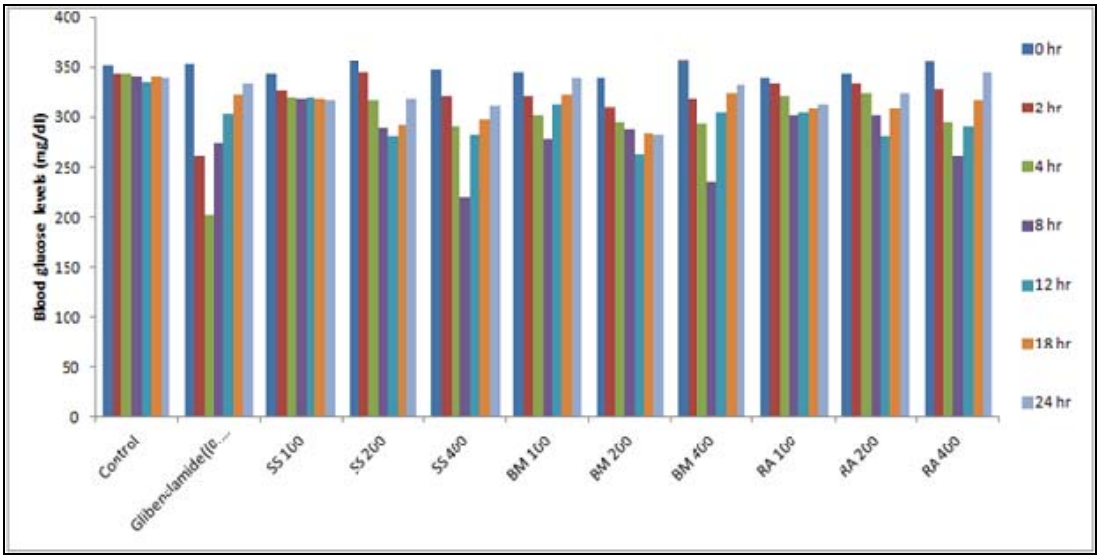

Fig 1: Effect of Methanol extracts of selected plants on blood glucose levels (mg/dl) in STZ induced diabetic rats at doses of 100,200 and 400 mg/kg b.w.

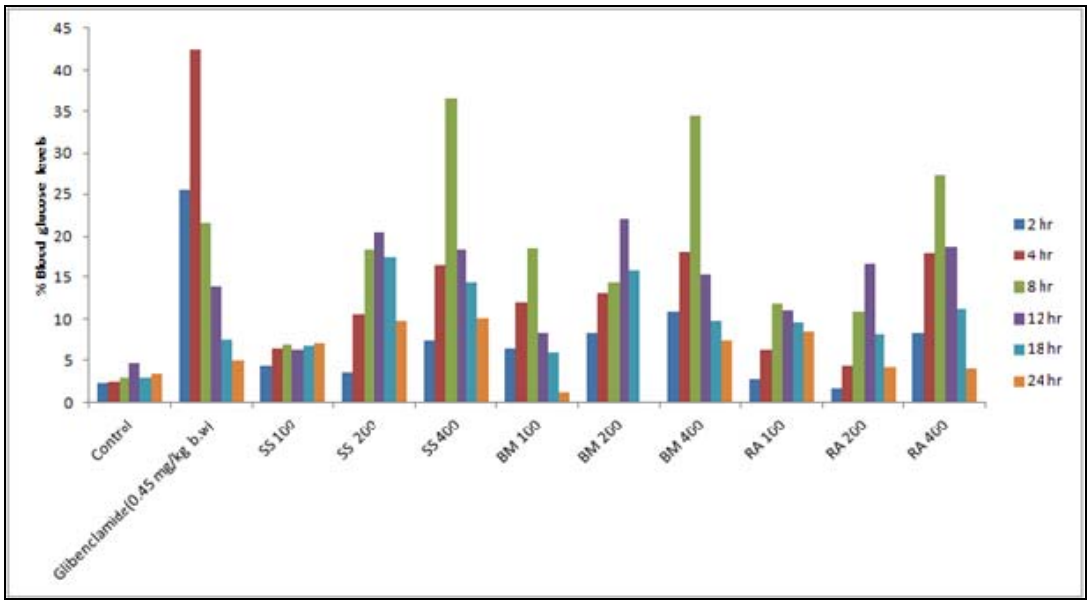

Fig 2: Effect of Methanol extracts of selected medicinal plants on percent decrease blood glucose levels in STZ induced diabetic rats

4. Conclusion

We finish from the behind results and argumentation on the plants of *Schrebera swietenoids*, *Barleria Montana* and *Rotula aquatica* harsh methanolic extracts has suited hypoglycemic potential. In the disclose investigation, we demonstrated that methanol commemorate of *Barleria montana* have happy effects on bracing the pull out of the fire of insulin by turning the spotlight on inhibition of serum blood levels in STZ possessed diabetic rats. Further studies are rightfully for the emptiness and anti-sepsis of bioactive compounds laid it on the line in the equivocate extracts and for explanation of their molecular mechanisms.

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