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Anti-Hyperglycemic potential of aqueous extract of leaves of *Solanum nigrum* Linn

Laxmidhar Maharana*[1], Snigdha Pattnaik[1], Durga M. Kar[1], Pratap K. Sahu[1], Sudam C. Si[1]

**1. School of Pharmaceutical Sciences, Siksha O Anusandhan University, Kalinga Nagar, Ghatikia, Bhubanesar, Dist: Khurda, Orissa, India*

Abstract

The hypoglycemic potential of aqueous (50, 100 mg/kg) extract of leaves of Solanum nigrum Linn (Solanaceae) was evaluated by normoglycemic rats followed by alloxan and glucose loaded hyperglycemic rats by single oral administration. The plant extract was subjected to the study of presence of different phytoconstituents by using standard qualitative chemical methods. The preliminary antioxidant potential of the extract was evaluated by spectrophotometric method, according to the Folin–Ciocalteu procedure for estimation of total phenolic content and calculated as gallic acid equivalents (GAE), whereas total flavonoid content was determined spectrophotometrically and expressed as quercetin equivalents. The related parameters like hematological, biochemical profiles including body weight variation study were also carried out in 30-days treated diabetic rats. The study report showed that the plant extract significantly ($p<0.01$ to $p<0.001$) reduces blood glucose level both in normoglycemic and hyperglycemic rats induced by alloxan and oral glucose loaded methods till the end of 10h and 4h respectively during the course of experiment. The preliminary phytochemical study report revealed that the test extract contain carbohydrate, poly peptides, saponin, tannins, alkaloids, flavonoids, terpinoids, coumarin; as phytoconstituents, whereas the Total phenolic and Total flavonoid content of the extract registered 33.83 µg of pyrocatechol equivalent /500mg , 5.86 mg equivalent of quercetin /gm respectively. The hematological, biochemical and loss of body weight study report are in good agreement with the activity and safety profile of the plant extract. The experimental results of the above studies indicate that the aqueous extract of the leaves of Solanum nigrum Linn endowed with potential anti-hyperglycemic activity presumably due to free radical scavenging potential of the test extract.

Key words: *Solanum nigrum linn.*, Alloxan, Hypoglycaemic, Glibenclamide, Insulin

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INTRODUCTION

The selection of scientific and systematic approach for the biological evaluation of plant products based on their use in the traditional systems of medicine forms the basis for an ideal approach in the development of new drugs from plants. Numbers of scientific and popular literatures has reported more than 1200 plants as hypoglycemic agents [1, 2], as plant drugs are frequently considered to be less toxic with lesser or rare side effects than those of synthetic ones [3]. *Solanum nigrum* Linn. (Solanaceae) commonly known as Black Berried Nightshade is a fairly common herb or short-lived perennial shrub, found in many wooded areas, as well as disturbed habitats. (*Solanum nigrum* plant profile, New South Wales Flora Online), distributed throughout India, Ceylon and all temperate and tropical regions of the world [4] . The leaves are known to be used to treat headache & diseases of nose [4], ringworm [5], heart & liver ailments, wounds & burns [6] , toothache [7]. The ethnomedical information reveals that the juice of dried leaves of *Solanum nigrum* is used for lowering blood sugar level [8]. Further, the aqueous extract of dried leaves is used for its antiviral [9], antipyretic, anticonvulsant, sedative, antimalarial, antispasmodic & diaphoretic [10], molluscicidal [11], anti-bronchitis & anti-gastralgia [12] activities. The *Kondh* tribes of Orissa, India use the hot aqueous extract of the fruits and leaves as a folk medicine for the treatment of diabetes mellitus. The leaves are reported to contain several constituents Quercitrin, Hyperoside [13], Sitosterol, Solamargine, Stigmastrol, Campesterol, Cholesterol [14], Solasodine [15]. The present study focus on the scientific validation of the traditional use of plant leaves for control of diabetes.

MATERIALS AND METHODS

Experimental

Plant material

The mature entire plant was collected from the place named as Konark in the state of Orissa and authenticated by Dr. A. K. Pradhan, Taxonomist of the Department of Botany, PPD Mahavidyalaya, Tigiria,Cuttack Orissa. A voucher specimen has been preserved in the institution herbarium of School of Pharmaceutical Sciences, Siksha ‘O’ Anusandhan University for future reference. After due authentication, fresh matured leaves were collected from well-grown plants at the same place and cleaned thoroughly to remove adherent from the leaves under running tap water. The cleaned leaf materials were subsequently dried under shade. The shade dried leaves were powdered in an electrical grinder and passed under sieve No. 40 to get coarse powder of the leaves for further study.

Preparation of extract

The powdered leaf materials were defatted using petroleum ether as solvent. The defatted powdered plant material (550 g) was refluxed with 1500 ml of distilled water for 48h followed by filtration and the filtrate was concentrated under vacuum. A dark brown sticky residue was obtained with yield value 21.52% (w/w).

Animals

The healthy Wistar albino rats, weighing 150–200g body weight of either sex were selected and housed in acrylic cages in standard laboratory conditions and were fed standard rodent diet with water *ad libitum*. The experiments on animals were conducted in accordance with the standard

experimental procedure and the animals were used as per the experimental protocol duly approved by the Institutional Ethical Committee of the School of Pharmaceutical Sciences, Siksha 'O' Anusandhan University, Bhubaneswar, Orissa with registration No. IAEC 1171/C/08/CPCSEA.

Screening for antidiabetic activity

The Screening for antidiabetic activity was conducted as per the method described by Dash et al. [16]. The test samples were suspended in 25% Tween 20 in distilled water. Glibenclamide (2.5 mg/kg) was used as reference control during the study. All the test samples were administered through oral route.

Study on normoglycaemic animals

The animals were fasted for 18 h, but were allowed to free access of water during course of the experiment. At the end of the fasting period, taken as zero time (0 h), blood was withdrawn (0.1 ml) from the tip of the tail of each rat under mild ether anaesthesia. Plasma was separated following centrifugation the glucose was estimated by GOD/POD method using Glucose estimation kit from M/s. Sigma Diagnostics (India) Pvt. Ltd., Baroda, India. The normal rats were then divided into four groups of six animals each. Group I served as solvent control and received only vehicle (2 ml/kg) through oral route, Group II received glibenclamide (2.5 mg/kg) and served as reference control. Groups III and IV received the test extract at a dose of 50 and 100 mg/kg, respectively, through oral route. All the test samples at 50, 100 mg/kg dose levels were administered in a similar manner. Blood glucose levels were examined after 1, 2, 4, 6, 8 and 10 h of administration of single dose of test and control samples.

Study on alloxan induced diabetic animals

The acclimatized animals were kept fasting for 24 h with water *ad libitum* and injected intraperitoneally a dose of 150 mg/kg of alloxan monohydrate in normal saline. After 1 h, the animals were provided feed *ad libitum*. The blood glucose level was checked before alloxanisation and 24 h after alloxanisation as above. Animals were considered diabetic when the blood glucose level was raised beyond 200 mg/100 ml of blood. This condition was observed at the end of 72 h after alloxanisation. The animals were segregated into four groups of six rats in each. Group I served as solvent control and received only vehicle (2 ml/kg) through oral route. Group II received glibenclamide (2.5 mg/kg). Groups III and IV received the test extract at doses of 50 and 100 mg/kg in a similar manner. Blood glucose level of each rat was estimated at 1, 2, 4, 6, 8 and 10 h, respectively.

Study on glucose loaded hyperglycaemic animals

The selected groups of animals were ingested with glucose (1 g/kg) in distilled water. The test and standard drug treatment were made as per the above manner and blood glucose level was measured at 0, 0.5, 1, 2, and 4 h interval respectively.

Qualitative phytochemical analysis

The presence of phytoconstituents in the test extract was carried out by standard chemical methods prescribed in Trease and Evans [17].

Determination of total phenolic content

Total soluble phenolic in the aqueous extract of Solanum nigrum was determined with Folin Ciocalteu reagent using pyrocatechol as a standard [18]. Briefly, 0.1 ml of extract solution (contain 1000 µg extract) in a volumetric flask was diluted in distilled water (46ml). About 1 ml of Folin Ciocalteu reagent was added and the content of the flask was mixed thoroughly. After 30 min, 3 ml of sodium bicarbonate was added, and then the mixture was allowed to stand for 2h with intermittent shaking. The absorbance was measured at 760 nm. The concentration of total phenolic content in the extracts was determined as microgram of pyrocatechol equivalent by using an equation that was obtained from standard pyrocatechol graph. The equation calculating the pyrocatechol was: Absorbance = 0.001 x pyrocatechol (µg) – 0.003

Determination of total flavonoid content

Aluminium chloride colorimetric method was used for flavonoid content determination [19]. Each extract (0.5 ml of 1 : 10 g/ml) in methanol was mixed with 1.5 ml of methanol, 0.1 ml 10 % aluminium chloride, 0.1 ml of 1 M potassium acetate and 2.8 ml of distilled water. It remained at room temperature for 30 min; the absorbance of the reaction mixture was measured at 415 nm. The calibration curve was prepared by preparing quercetin solutions at concentrations 12.5 to 100 µg/ml in methanol.

Sub acute toxicity studies

The sub acute toxicity study on alloxnised diabetic rats was conducted for 30-days under the treatment of test extract, standard drug and solvent in a similar manner as per the above experiments. The parameters considered for the study are body weight variation test by measuring weight of the animals by standard weighing method using balance and study of hematological profile along with serum biochemical changes by following standard laboratory methods. The blood sample under study was collected on 30^{th} day by sacrificing the animals under anesthetic condition and serum was separated out by freezed centrifugation using a high speed Remi centrifuge. The body weight of the animals were recorded on 0, 10, 20 and 30 days of the experiment. The haematological studies include total RBC count, total WBC count, clotting time and differntial count and % Hb [20]. Other biochemical/serum profile studies include ASAT, ALAT, ALP, TB, DB, albumin, total protein, globulin were also performed by using standard established procedures [21].

Statistical analysis

All the results were analysed statistically using one-way analysis of variance (ANOVA) followed by Dunnet's t-test. A *p*-value less than 0.05 is considered significant. All the results are expressed as Mean ± S.E.M for six animals in each group.

RESULTS AND DISCUSSION

The preliminary phytochemical investigation report indicates that the aqueous extract of *S. nigrum* contains carbohydrates, poly peptides, saponins, tannins, alkaloids, flavonoids, coumarin, terpinoids, sterols as phytoconstituents but devoid of glycosides, fats and oils and steroidal constituents. The experimental results of the effect of aqueous extract of *Solanum nigrum* leaves in normoglycemic rats (Table 1) showed that blood glucose levels decrease significantly ($p<0.01$) with effect from 6h onwards till the end of 10h, in case of standard and test extract

treated group, while the % reduction of glucose levels calculated as 43 (standard drug), 40 to 21 (test extract) respectively. The results of the normoglycemic model showed that the test extract have dose dependent hypoglycemic effect. The perusal of Table No. 2 concerning antidiabetic activity in alloxan induced diabetic rats showed that the test extract in both dose levels, reduces the blood glucose significantly ($p<0.01$) starting from 2h to the end of 10h of the study in a dose dependent manner, while the standard drug, glibenclamide showed similar effect during the course of the experiment. However the percent decrease of blood sugar at the end of 10h calculated as 46 to 55%, while standard drug showed 72% at the same time. The study of oral glucose loaded hyperglycemic model also significantly reduces the blood glucose level ($p<0.01$) in dose level of 100 mg/kg (38%), while standard drug registered 48% with statistical significant reduction till the end of 4 h (Table 3). The statistical significance of ANOVA in showed significant reduction of blood glucose with $p<0.05$ to $p<0.001$.

It is generally accepted that alloxan treatment causes permanent destruction of β-cells and impairment of renal function; and sulfonylureas are known to lower the blood glucose level by stimulating β-cells to release insulin [22]. However, the statistically significant anti-hyperglycemic as well as hypoglycemic activities shown by the aqueous extract of *S. nigrum* leaves in both single dose treated normoglycaemic and hyperglycaemic models might suggest that the said effect be due to extra-pancreatic and extra-intestinal action of the test extract [23]. And the decreased activity in glucose level in OGTT might be, due to a decrease in the rate of initial glucose absorption when plant fiber is given orally with glucose [23].

Phenolic compounds such as tannins, flavonoids and phenolic acids are considered to be the major contributors to the antioxidant capacity of plants. All phenols and particularly flavonoids are effective antioxidants because they donate electrons to radicals and break the radical chains. Phenolic compounds have been shown to exert a wide range of biological activities including scavenging ROS [24]. Since the total phenols and total flavonoids contents of aqueous extract of leaves of *S.nigrum* are found to 33.83 µg of pyrocatechol equivalent /500mg and 5.86 mg equivalent of quercetin /gm, which is quantitatively a greater value, hence it is presumed that, the antioxidant potential of the extract may plays a significant role for anti hyperglycemic potential of the plant extract.

Table 1: Hypoglycemic activity of aqueous leave extract of *Solanum nigrum* (ALSN) in single dose treated normoglycemic rats in oral route.

Groups & Treatment	Blood Glucose Levels (mg/dl)							
	0 h	1 h	2 h	4 h	6 h	8 h	10 h	%age decrease at 10hrs
I. Solvent Control (Tween + Water)	103.5 ± 2.71	101.5 ± 3.88	104.16 ± 4.33	102.66 ± 4.26	102.83 ± 3.23	104.66 ± 3.04	103.16 ± 2.84	1.93
II. Glibencamide (2.5mg/kg)	101.33 ± 6.45	99.5 ± 5.77	94.66 ± 4.53	89.33 ± 3.63[a]	79.16 ± 2.56[c]	67.33 ± 2.90[c]	57.16 ± 3.26[c]	43.59
III. ALSN (50mg/kg)	108.83 ± 4.03	106.16 ± 3.91	105. 83 ± 3.57	98.83 ± 4.05	95.16 ± 4.33	87.33 ± 3.02[b]	85.83 ± 3.04[b]	21.13
IV. ALSN (100mg/kg)	98.66 ± 3.56	96.83 ± 3.16	94.16 ± 2.61	91.16 ± 2.84	87.66 ± 3.01[b]	70.33 ± 3.63[c]	59.5 ± 3.21[c]	39.69
F (3,20)	0.95	0.84	2.56	2.84	9.15**	24.17**	23.45**	-

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test.
(F-value denotes statistical significance at *p<0.05, **p<0.01)
(t-value denotes statistical significance at [a]p<0.05, [b]p<0.01 and [c]p<0.001 respectively, in comparison to group-I).

Table 2: Anti-diabetic activity of aqueous leave extract of *Solanum nigrum* (ALSN) in single dose treated alloxan induced hyperglycemic rats in oral route.

Groups & Treatment	Blood Glucose Levels(mg/dl)							
	0 h	**1 h**	**2 h**	**4 h**	**6 h**	**8 h**	**10 h**	**%age decrease at 10hrs**
I. Solvent Control (Tween + Water)	271.66 ± 10.01	272.5 ± 5.03	282.5 ± 4.85	278.16 ± 9.16	287.33 ± 13.09	269.66 ± 14.14	268.83 ± 9.81	1.04
II. Glibencamide (2.5mg/kg)	282.66 ± 2.62	225.16 ± 4.04^{b}	176.16 ± 5.22^{c}	114.33 ± 6.62^{c}	105.83 ± 7.12^{c}	88.33 ± 3.27^{c}	78.83 ± 7.21^{c}	72.11
III. ALSN (50mg/kg)	266.26 ± 9.15	257.16 ± 7.65	243.83 ± 6.96^{b}	178.66 ± 10.93^{c}	159.33 ± 7.63^{c}	146.33 ± 6.25^{c}	141.5 ± 5.47^{c}	46.85
IV. ALSN (100mg/kg)	283.83 ± 10.08	254.33 ± 11.9	214.83 ± 8.51^{c}	158.83 ± 7.76^{c}	141.83 ± 10.25^{c}	137.66 ± 10.83^{c}	126.83 ± 7.91^{c}	55.31
F (3,20)	1.00	6.43**	47.12**	62.26**	64.77**	64.27**	109.16**	-

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test.
(F-value denotes statistical significance at *p<0.05, **p<0.01)
(t-value denotes statistical significance at ^{a}p<0.05, ^{b}p<0.01 and ^{c}p<0.001 respectively, in comparison to group-I).

Table 3: Effects of single dose treatment of aqueous leave extract of *Solanum nigrum* (ALSN) in glucose loaded hyperglycemic rats in oral route.

Groups & Treatment	Blood Glucose Levels (mg/dl)					
	0 h	**0.5 h**	**1 h**	**2 h**	**4 h**	**%age decrease at 4hrs**
I. Solvent Control (Tween + Water)	144.16 ± 5.06	135.16 ± 4.65	129.16 ± 2.88	121.5 ± 1.72	113.5 ± 2.47	21.26
II. Glibencamide (2.5mg/kg)	157.83 ± 6.53	134.66 ± 5.25	117.00 ± 4.57	95.16 ± 2.03^{c}	82.33 ± 2.31^{c}	47.83
III. ALSN (50mg/kg)	151.66 ± 3.84	139.66 ± 3.11	122.33 ± 5.37	116.5 ± 5.03	115.83 ± 2.22	23.62
IV. ALSN (100mg/kg)	158.66 ± 5.73	136.5 ± 5.90	116.33 ± 5.25	105.5 ± 4.25^{b}	98.66 ± 3.56^{b}	37.81
F (3,20)	1.55	0.21	1.65	10.90**	32.92**	-

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test.
(F-value denotes statistical significance at *p<0.05, **p<0.01)
(t-value denotes statistical significance at ^{a}p<0.05, ^{b}p<0.01 and ^{c}p<0.001 respectively, in comparison to group-I).

Diabetes mellitus causes failure to use of glucose for energy which leads to increased utilization and decrease storage of protein responsible for reduction of body weight essentially by depletion of body proteins [25]. The percentage losses in the body weight of the animals were recorded on 0^{th}, 10^{th}, 20^{th} & 30^{th} day of the experiment and shown in Table 4. The % loss of body weight during 30-days study in diabetic rats under treatment of test and standard drug, showed that, there are significant ($p<0.05$ to $p<0.001$) recovery of body weight when compared with solvent treated diabetic rats. The property of the test extract to recover the body weight of animals suggesting the extra pancreatic action of the extract and might be contributed by increased utilization of glucose by the tissues.

Table 4: Effect of aqueous leave extract of *Solanum nigrum* (ALSN) on percentage loss in body weight in multi-dose treated alloxan induced diabetic rats.

Groups & Treatment	Percentage loss in body weight			
	0^{th} day	**10^{th} day**	**20^{th} day**	**30^{th} day**
I. Solvent Control (Tween + Water)	27.16 ± 2.25	31.5 ± 2.12	34.66 ± 2.27	37.83 ± 3.15
II. Glibencamide (2.5mg/kg)	26.16 ± 3.75	19.5 ± 2.36^{b}	9.66 ± 1.05^{c}	5.5 ± 0.71^{c}
III. ALSN (50mg/kg)	29.33 ± 2.76	24.33 ± 1.94^{a}	14.5 ± 1.23^{c}	11.5 ± 1.23^{c}
IV. ALSN (100mg/kg)	23.16 ± 1.40	20.33 ±0.91^{b}	12.16 ± 1.01^{c}	8.83 ± 1.07^{c}
F (3,20)	0.91	8.15**	59.35**	66.74**

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test, (F-value denotes statistical significance at *$p<0.05$, **$p<0.01$), (t-value denotes statistical significance at $^{a}p<0.05$, $^{b}p<0.01$ and $^{c}p<0.001$ respectively, in comparison to group-I).

The hematological profile of diabetic rats under 30-days treatment of the extract, glibenclamide and solvent, once daily in both dose levels (Table 6) showed that, there is a no marked alteration in the values of tested haematological parameters, in the extract treated group except value of neutrophil, when compared with diabetic control, suggesting safetyness of the test extract on long term use.

Table 5: Effect of aqueous leave extract of *Solanum nigrum* (ALSN) on serum haematological parameters in alloxanised rats on 30th day of study.

Groups and Treatment	Serum Haematological parameters								
	RBC (millions/ml)	**WBC (1000/ml)**	**Hb (g/dl)**	**Clotting time (min.)**	**Neutrophil (%)**	**Eosinophil (%)**	**Basophil (%)**	**Lymphocyte (%)**	**Monocyte (%)**
I. Normal	4.19 ± 1.35	6.65 ± 1.2	9.86 ± 1.05[a]	1.08± 0.18	28.5 ± .40[a]	3.5 ± 0.84[a]	00	68.16 ± 4.39	1.8 ± 0.32[a]
II. Solvent Control (Tween+Water)	2.41 ± 1.15	7.38 ± .14	14.75 ± 1.71	1.61± 0.19	20.5 ± 2.45	6.5 ± 1.25	00	72.33 ± 5.13	4.1 ± 0.77
III. Glibencamide (2.5mg/kg)	4.05 ± 1.07	6.88 ± .37	13.41 ± 0.96	1.01± 0.14	26.91±1.44	4.66 ± 0.76	00	67.33 ± 7.12	2.0 ± 0.82[a]
IV. ALSN (50mg/kg)	3.15 ± 0.91	5.35 ± .06	11.16 ± 1.19	1.21± 0.22	20.75± .63	2.83 ± .90[a]	00	72.33 ± 9.17	2.3 ± 0.74
V. ALSN (100mg/kg)	3.45 ± 0.87	6.58 ± .53	14.04 ± 1.72	1.18 ± 0.22	26.08±2.15	3.33 ± .76[a]	00	69.16 ± 8.52	2.2 ± 0.64
F (4, 25)	0.43	0.34	2.24	1.38	2.67*	2.51	--	0.10	1.87

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test. (F-value denotes statistical significance at *p<0.05, **p<0.01), (t-value denotes statistical significance at [a]p<0.05, [b]p<0.01 and [c]p<0.001 respectively, in comparison to group-II.)

The serum biochemical investigation report (Table 6) containing estimation of marker enzyme in alloxanised animals showed, the enzymes like ASAT, ALAT and ALP which are considered to be good indices of liver and kidney damage [26] are significantly reduced by the extract and therefore it may be presumed that the extract protects cellular damage. The other serum biochemical parameters like Total bilirubin, direct bilirubin, Albumin, Total protein, globulin were found insignificant different when compared with reference animal group.

Table 6: Effect of aqueous leave extract of *Solanum nigrum* (ALSN) on serum biochemical parameters in alloxanised rats on 30th day of study.

Groups and Treatment	Serum Biochemical parameters							
	ASAT (u/l)	**ALAT (u/l)**	**ALP (u/l)**	**TB (mg/dl)**	**DB (mg/dl)**	**Albumin (gm/dl)**	**Total Protein (gm/dl)**	**Globulin (gm/dl)**
I. Normal	22.83 ± 2.42^{c}	28.91 ± 2.41^{c}	105.38 ± 11.12^{c}	0.91 ± 0.14	0.25 ± 0.07	3.71 ± 1.00	6.61 ± 0.64	1.96 ± 0.34^{a}
II. Solvent Control (Tween + Water)	42.08 ± 2.80	57.16 ± 4.14	258.5 ± 15.64	1.55 ± 0.38	0.31 ± 0.03	5.33 ± 1.20	4.38 ± 0.68	1.01 ± 0.25
III. Glibencamide (2.5mg/kg)	24.2 ± 3.14^{c}	29.41 ± 3.58^{c}	135.25 ± 13.15^{c}	0.74 ± 0.12^{a}	0.22 ± 0.01	4.31 ± 1.01	5.95 ± 0.71	1.26 ± 0.26
IV. ALSN (50mg/kg)	39.75 ± 2.50	42.41 ± 4.08^{a}	143.16 ± 10.84^{c}	1.30 ± 0.11	0.41 ± 0.04	5.08 ± 1.15	4.26 ± 0.69	0.73 ± 0.13
V. ALSN (100mg/kg)	32.25 ± 2.30^{a}	40.78 ± 4.12^{a}	138.91 ± 11.01^{c}	1.22 ± 0.17	0.38 ± 0.04	4.53 ± 1.15	4.81 ± 0.93	1.11 ± 0.24
F (4, 25)	10.88**	9.62**	22.35**	2.22	3.10*	0.33	1.93	3.17*

Values are expressed in MEAN ± S.E.M of six animals. One Way ANOVA followed by Dunnet's t-test. (F-value denotes statistical significance at *p<0.05, **p<0.01), (t-value denotes statistical significance at ^{a}p<0.05, ^{b}p<0.01 and ^{c}p<0.001 respectively, in comparison to group-II).

CONCLUSION

In this study, several animal models and in vitro assay were applied to evaluate the antihyperglycemic potential of S. nigrum, leaves aqueous extract. Results indicate that the leaf of S. nigrum endowed with antihyperglycemic activity presumably due to the antioxidant potential of the leaf of the plant.

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REFERENCES

[1] Marles RJ and Farnsworth NR, *Phytomedicine*, **1995**, 2 (2): 137-189.
[2] Wang HX and Ng TB, *Life Sci*, **1999**, 65: 2663-2677.
[3] Pari L and Umamaheswari J, *J Ethnopharmacol*, **2000**, 14: 136-138.
[4] Kirtikar KR and Basu BD, Indian Medicinal Plants. Vol. III, Book Publisher, India: **1999**, 1748-1751.
[5] Rana TS and Datt B, *India. Int J Pharmacog*, **1997**, 35 (5): 371-374.
[6] Dafni A and Yaniv Z, *J Ethnopharmacol*, **1994**, 44 (1):11-18.
[7] Bocek BR, *Econ Bot*, **1984**, 38 (2):240-255.
[8] Villasenor IM and Lamadrid MRA, *J Ethnopharmacol*, **2006**, 104 (1-2):129-131.
[9] Roychoudhury R and Basu PK, *Indian J Exp Biol*, **1983**, 21 (4):212-215.
[10] Adesina SK, *Fitoterapia*, **1982**, 53: 147-162.
[11] Kloos H, Thiongo FW, Ouma JH and Butterworth AE, *J Trop Med Hyg*, **1987**, 90 (4):197-204.
[12] Darias V, Bravo L, Barquin E, Herrera DM and Fraile C, *J Ethnopharmacol*, **1986**, 15 (2):169-193.
[13] Nawwar MAM, El-Mousallamy AMD and Barakat HH, *Phytochemistry*, **1989**, 28 (6):1755-1757.
[14] Bhatt PN and Bhatt DP, *J Nat Prod*., **1984**, 47 (3): 426-432.
[15] Verbist JF, Monnet R and Dobremez JF, *Plant Med Phytother*., **1977**, (11): 40-46.
[16] Dash GK, Suresh P and Ganapaty S, *Journal of Natural Remedies*, **2001**, 1: 105–110.
[17] Trease GE and Evans MC, Text book of Pharmacognosy. 12th edition. Balliere, Tindall, London, **1983**, p:343-383.
[18] Slinkard K and Singleton VL, *Am. J. Enol. Vitic*., **1977**, 28, 49-55.
[19] Chang C, Yang M, Wen H and Chern J., *J. Food Drug Anal*., **2002**, 10, 178-182.
[20] Ghai CL, A Text Book of Practical Physiology. Jaypee Brothers Medical Publishers, 6th ed. New Delhi, **2004**, p: 130-175.
[21] Sood R, Clinical chemistry In: Medical laboratory technology, methods and interpretations, Jaypee brothers' medical publishers, India 5th ed., **1999**, pp. 173-404.
[22] Pari L and Maheswari J, *J Ethnopharmacol*, **1999**, 68:321-325.
[23] Day C, Catwright T, Provost J and Bailey CJ, *Planta Medica*, **1990**, 56: 426–429.
[24] Frankel EN, Natural phenolic antioxidant and their impact on health, in: Packer L, Hiramatsu M and Yoshikawa T, (Eds.), Antioxidant food supplements in human health, Academic press, San Diego, **1999**, pp: 385-392.
[25] Guyton AC and Hall JE, Guyton and Hall Text Book of Medical Physiology. 10th ed. Elsevier, a division of Reed Elsevier India Pvt. Ltd., New Delhi, **2000**, pp. 894–897.
[26] Martin DW, Mayes PA and Rodwell YW, Transaminases In: Harper's Review of Biochemistry, Lange Medical 18th ed., **1981**, pp:61.