Research Article

Evaluation of Antidiabetic and Hypolipidemic Activities of Ethanolic Extract of *Portulaca oleracea* (Whole Plant) in Alloxan Induced Diabetic Rats

Sabeeha Shafi*, Nahida Tabassum
Department of Pharmaceutical Sciences, University of Kashmir, Hazratbal, Srinagar, Kashmir, Jammu & Kashmir, India.
***Corresponding author's E-mail:** sabeeha_shafi@yahoo.com

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ABSTRACT

Portulaca oleracea commonly called as Nunar in Kashmiri, Common Purslane in English and Lunia in Hindi is a weed that is cosmopolitan in nature and grows in warm temperate, tropical and subtropical regions of the world. Many chemical constituents have been isolated from this plant which has shown a number of pharmacological activities of this plant. These pharmacological activities include antifungal, analgesic, anti-inflammatory, gastric antiulcerogenic, bronchodilatory and anti-tumour activities. Very little work has been done on anti-diabetic and hypolipidemic activities. The present study was to ascertain the chemical constituents of this plant and evaluate the antidiabetic and hypolipidemic potential of the hydro-alcoholic extract of the plant. The study has revealed that the hydro-alcoholic extract of whole plant of *Portulaca oleracea* has various chemical constituents and significant anti-diabetic and hypolipidemic activities.

Keywords: *Portulaca oleracea*, antidiabetic, hypolipidemic.

INTRODUCTION

Diabetes mellitus is a lifestyle metabolic disease in the world today. It affects at least 15 million people. It is associated with long term complications, including retinopathy, nephropathy, neuropathy and angiopathy and several others.[1] Being a multifactorial disease characterized by hyperglycemia, lipoprotein abnormalities, raised basal metabolic rate, defect in reactive oxygen species scavenging enzymes and high oxidative stress induced damage to pancreatic beta cells. It has been ranked third among the leading causes of death when its fatal complications are taken into account. In India alone there are more than 4.00 crore diabetics and the number is going to be around 9.00 crore by 2030. Over 7.20 lakh Indians die every year due to diabetes. It has been seen that people with diabetes are 2-4 times more likely to develop heart diseases.[2]

Various efforts have been taken to understand and manage diabetes mellitus because the disease and disease related complications are increasing day by day. India has 45,000 plant species and many of them have medicinal properties.[3-12] About 800 plant species have shown anti-diabetic activity. People have shown great demand for plant products due to low cost, easy availability and lesser side effects. For this plant materials are continuously scrutinized and explored for their effect as antidiabetic agents.

Portulaca oleracea Linn, belonging to Family Portulacaceae (Purslane family) is commonly called as Common Purslane/Purslane in English, as Kurfa in Mumbai, as Loni, Ghol in Gujrati, as Kursa, Chhota Lunia in Hindi, as Lonak in Punjabi and as Nunar in Kashmiri is a cosmopolitan weed in warm temperate, tropical and subtropical regions of the world. It grows along waste lands and in cultivated gardens in Srinagar. It also contains carboxylic acids, gums , fatty acids, beta-carotene and volatile oil and Portuloside A**,** a monoterpene glucoside and phenolic alkaloids.[13-17] In folk medicine it is reported that it can be used as a salad, cooked like soups. It is used to treat burns earache, insect stings, inflammation, skin sores, ulcers, pruritis (itching skin) eczema and abscesses. It is used in treatment of cardiovascular disorders, dysuria, haematuria, gonorrhoea, dysentery, sore nipples and ulcers of mouth. It is used as blood purifier. Roasted seeds are reported to be diuretic and anti-dysenteric. Omega 3–fatty acids present are used in the production of compounds that effect blood pressure clotting, the immune system, lower cholesterol (LDL) and prevent certain cancers and control coronary spasms. They have positive effect on brain and in such conditions as depression, bipolar disorder, alzhemiers disease, schizophrenia, hyperactivity and migrane. Reported Pharmacological Activities include antifungal, antibacterial, analgesic, anti-inflammatory, gastric antiulcerogenic, bronchodilatory, skeletalmusclerelaxant, antihypertensive, neuropharmacological, wound healing, antioxidant, antifertility and antitumour activities.[18-30] Therefore, with the reference to traditional and reported uses, the present study was undertaken to investigate the antidiabetic and hypolipidemic activity of this plant and give a scientific rational for its use.

MATERIALS AND METHODS

Plant Material

The plant of *Portulaca oleracea* was collected from specific areas, Shalimar area of the district, Srinagar. It was collected during the months of April to June and authenticated by a plant taxonomist in the Centre of

Plant Taxonomy, University of Kashmir, Srinagar. A sample of the plant material was deposited in the herbarium of the Department of Taxonomy, University of Kashmir under voucher specimen number 1012(KASH) for future reference. This plant material was dried. It was kept in a well ventilated room with outside temperature ranging between 18 to 32°C.

Preparation of the extract

Portulaca oleracea (whole plant) was coarsely powdered and 500 gm of the material was macerated for 48 hrs with 50% ethanol, with occasional shaking. After 48 hrs, the ethanolic extract was filtered through Whatmans filter paper. This plant material was then macerated again with fresh 50% ethanol. The filtrate obtained from the first and the second maceration was then combined and the solvent was recovered. After the recovery of alcohol, the extract was then evaporated to dryness. The yield was noted. The extract of the plant was refrigerated at 4°C for future use in experimental studies.

Phytochemical Screening

The hydro-alcoholic extract obtained was subjected to qualitative tests for identification of different constituents like tannins, alkaloids, saponins, glycosides, terpenes, phenolics, flavonoids, carbohydrates, proteins and steroids, by using simple and standard qualitative methods described by Trease and Evans.[31-33]

Pharmacological Study[34]

Animals

Healthy albino rats of either sex were used during the study weighing about 180-210 g. These animals were procured from Central Animal House, IIIM (Indian Institute of Integrative Medicine) Jammu. They were housed in clean polypropylene cages. The rats were acclimatized for a period of 7 days, before initiation of experiment. Standard environmental conditions such as temperature ranging from 18 to 32°C, relative humidity (70%) and 12 hrs dark/light cycle were maintained in the quarantine. These animals were fed with rodent pellet diet (Ashirwad Industries) and water *ad-libitum* under strict hygienic conditions. All procedures were performed in accordance to CPCSEA guidelines after approval from the Institutional Animal and Ethics Committee (IAEC) of the Department of Pharmaceutical Sciences, University of Kashmir [No. F-IAEC (Pharm.Sc) APPROVAL].

Induction of diabetes

Alloxan monohydrate was used to induce diabetes. The chemicals used were of analytical grade and were acquired from commercial sources. A single dose (120mg/kg, b.w, i.p) of alloxan monohydrate in sterile saline was used for the induction of diabetes in rats after overnight fasting. After one hour of alloxan administration, the animals were fed standard pellets and water *ad libitum*. After 5 days of alloxan administration, animals showing blood glucose levels above 250 mg/dl were selected for the study. Hydro-alcoholic extract of *Portulaca oleracea* (PO) was administered at two dose levels 100 and 200 mg/kg b.w.

Experimental design

These rats were fasted overnight for 12 hrs. They were randomly divided into 5 groups of 6 rats per group. The various groups were:

Group I-Normal control and received only 0.2 ml of 2% aqueous gum acacia

Group II-Diabetic control and received only alloxan monohydrate and 2% aqueous gum acacia.

Group III-Alloxan monohydrate + Glibenclamide (10 mg/kg, p.o) and served as Standard Antidiabetic drug.

Group IV-Alloxan monohydrate + 50% Ethanolic extract of PO (100 mg/kg, p.o)

Group V-Alloxan monohydrate +50% Ethanolic extract of PO (200 mg/kg, p.o)

The treatment with hydro-alcoholic extract of *Portulaca oleracea* was started on the same day except normal control and diabetic control groups which received only 0.2 ml of 2% aqueous gum acacia for a period of 10 days.

Animals in all groups had free access to standard diet and water during this period. Body weight and blood glucose levels were estimated on 1st, 4th, 7th and 10th day of the treatment.

Sample Collection

The blood samples were collected by pricking the tail from overnight fasted rats and blood glucose levels were estimated using One Touch Ultra glucose strips (Johnson & Johnson Ltd) on 1st, 4th, and 7th day.

Estimation of biochemical parameters

On the 10th day of the experiment, blood was collected from overnight fasted rats under ether anesthesia by cardiac puncture. It was kept aside for 30 min for clotting. By centrifuging the same sample at 6000 rpm for 20 min, the serum was separated and was analyzed for blood glucose[35,36] total cholesterol[37], triglycerides,[38] HDL cholesterol[39] and LDL cholesterol[40].

Statistical analysis

All the values are expressed as mean ± SEM. The results were subjected to statistical analysis using one-way ANOVA followed by students t test. $p<0.001$ was considered very highly significant.

RESULTS

Phytochemical analysis

The phytochemical analysis of the extract showed the presence of alkaloids, flavonoids, glycosides, carbohydrates, tannins, terpenes, steroids, Proteins, saponins and phenolics

Table 1: Phytochemical Results of *Portulaca oleracea* (whole plant)

S. No	Phytoconstituents	Results
1	Tannins	+
2	Alkaloids	+
3	Saponins	+
4	Glycosides	+
5	Terpenes	+
6	Phenolics	+
7	Flavonoids	+
8	Carbohydrates	+
9	Proteins	+
10	Steroids	+

Antidiabetic activity: The blood glucose levels showed a highly significant decrease in groups III, IV and V ($p<0.01$) when it was compared to group II (Diabetic control). There was a highly significant increase in blood glucose levels seen in diabetic group as compared to normal control group I ($p< 0.01$). **(Table 2)**

Effect of hydro-alcoholic extract of *Portulaca oleracea* (whole plant) on biochemical parameters in alloxan induced diabetic rats.

The biochemical parameters showed significant results. The serum total cholesterol levels showed a highly significant decrease in groups IV and V ($p<0.01$) as compared to diabetic control (Group II). Serum triglyceride levels showed a highly significant decrease in groups IV and V ($p< 0.01$). HDL levels showed a non significant increase in groups III, IV and V. LDL levels showed a significant decrease in groups IV and V groups **(Table 3).**

Effect of hydro-alcoholic extract of whole plant of *Portulaca oleracea* on body weight in alloxan induced diabetic rats

The animals of Normal Control Group were found to be stable in their body weight but diabetic rats showed significant reduction in body weight after 10 days. ($p< 0.01$) Alloxan mediated body weight reduction was reversed by the hydro-alcoholic extract in dose dependent fashion 100 mg/kg and 200 mg/kg b.w showed a highly significant increase in body weight ($p <0.01$).

The effect of extract at 200 mg/kg on body weight of the animals was also found statistically significant. Results on body weight are shown (**Table 4).**

Table 2: Effect of hydro-alcoholic extract of *Portulaca oleracea* (whole plant) on fasting blood glucose level (mg/dl) in alloxan induced diabetic rats

Group	Treatment	1st day	4th day	7th day	10th day
I	Normal control 0.2 ml of 2% aqueous Gum acacia	85.07± 4.35	86.16±4.43	84.82±5.96	84.71±6.11
II	Diabetic control (Vehicle) 0.2 ml of 2% aqueous gum acacia	261.47±8.37	264.28±8.29	268.03±8.48	271.33±8.18***
III	Alloxan monohydrate + glibenclamide (10 mg/kg)	200.37±5.25	141.18±2.43	124.52±2.00	114.84±3.21***
IV	Alloxan monohydrate+ 50% Ethanolic extract (PO, 100 mg/kg)	203.38±4.04	147.87±2.30	144.56±2.56	142.82±2.76***
V	Alloxan monohydrate+50% Ethanolic extract(PO,200 mg/kg)	201.24±4.90	146.29±2.06	130.44±1.87	111.57±2.67***

Animal: Albino Rats; Alloxan: 120 mg/kg.i.p; Extract: p.o.; Values are Mean ±S.E.M n=6; ***P< 0.001 highly significant Groups III, IV, V vs Diabetic Control (Group II) and Group I vs Group II on 10th day

Table 3: Effect of 50% ethanolic extract of Portulaca oleracea (whole plant) on lipid profile in alloxan induced diabetic rats.

Group	Treatment	Serum total Cholesterol mg/dl	Serum triglyceride mg/dl	Serum HDL Cholesterol mg/dl	Serum LDL Cholesterol mg/dl
I	Normal control 0.2 ml of 2% aqueous gum acacia	85.25±8.51	80.71±9.38	24.54±6.49	52.03±3.21
II	Diabetic control 0.2 ml of 2% aqueous gum acacia	218.15±23.79	194.56±14.99	21.11±1.45	89.59±13.82
III	(Alloxan monohydrate +standard drug glibenclamide 10 mg/kg)	206.35±6.11*	184.30±9.68*	29.03±3.16*	85.39±7.24*
IV	Alloxan monohydrate + Ethanolic extract (PO,100 mg/kg)	196.80±25.95**	140.58±18.62**	26.63±5.39*	143.14±19.38**
V	Alloxan monohydrate +Ethanolic extract (PO,200 mg/kg)	101.85±19.17**	97.32±12.15***	29.04±7.67*	60.06±12.46**

Animal: Albino Rats; Alloxan: 120 mg/kg.i.p; Extract: p.o.; Value are Mean ±S.E.M: n=6; * p> 0.05 non significant; **p< 0.05 significant; ***P< 0.01 highly significant; Groups III, IV, V vs Diabetic Control (Group II) and Group I vs Group II on 10th day

Table 4: Effect of 50% ethanolic extract of *Portulaca oleracea* (whole plant) on average body weight (g) in alloxan induced diabetic rats

Group	Treatment	Average Body weight of the animal (g)			
		1st day	4th day	7th day	10th day
I	Normal control 0.2 ml of 2% aqueous gum acacia	228.85±6.03	229.88±6.66	233.00±7.07	222.05±4.75
II	Diabetic control 0.2 ml of 2% aqueous gum acacia	180.58±3.66	163.86±2.08	152.21±3.12	124.76±2.35
III	Alloxan monohydrate + standard drug glibenclamide (10 mg/kg)	183.83±3.34	173.85±3.37	152.98±4.44	137.50±3.54***
IV	Alloxan monohydrate + Ethanolic extract (PO,100 mg/kg)	167.20±2.83	167.00±1.64	163.12±2.73	145.50±2.74***
V	Alloxan monohydrate + Ethanolic extract (PO,200 mg/kg)	156.37±3.74	153.98±3.24	148.15±3.41	150.61±4.05***

Animal: Albino Rats; Alloxan: 120 mg/kg. i.p; Extract: p.o.; Value are Mean ± S.E.M: n=6; * p> 0.05 non significant; **p< 0.05 significant; ***P< 0.01 highly significant; Groups III, IV, V vs Diabetic Control (Group II) and Group I vs Group II on 10th day

DISCUSSION

The pancreas is the primary organ of the body involved in sensing the organism's dietary and energetic states through glucose concentration in the blood and in response to elevated blood glucose, insulin is secreted. Alloxan has been the usual substance used for the induction of diabetes mellitus apart from streptozotocin. It has a destructive effect of the beta cells of the pancreas Alloxan causes a massive reduction in insulin release by the destruction of beta-cells of the islets of Langerhans thereby inducing hyperglycemia[41] Insulin deficiency leads to various metabolic alterations in the animals viz increased blood glucose and increased lipid profile. Diabetes mellitus is a metabolic disorder characterized by resistance in the action of insulin, insufficient insulin recreation or both. It is one of the most common diseases of the world. Type II diabetes in young has increased 30 fold over the last 20 years concomitant with increase in obesity. Studies have revealed that all incidences of diabetes in this young age group is 2.5% and alarmingly 25% of their young adults have abnormalities of blood glucose.[1]

Herbal plants have received greater attention as an alternative to conventional therapy. The demand for these remedies has currently increased. Experimental screening method is imperative in order to establish the safety and efficacy of traditional and herbal products and also to set up the active components of the herbal products.

The Indian indigenous drugs have great importance both from professional and economic point of view.[3-12] A large number of plants have been reported to possess anti-diabetic activity e.g., *Aconitum napeilus, Aloe vera, Carum carvi, Cichorium intybus, Allium cepa, Aralia cachemirica, Allium sativum, Momordia charantia.*

Rats weighing in the range of 180-210 g were procured from IIIM Jammu and kept in polypropylene cages under uniform conditions of food, water, temperature and degree of nursing care. It was ensured that the animals were in good health. These animals were free from diseases. Male and female animals were kept in separate cages so that there was no interference in evaluation of biochemical parameters during the period of study. The temperature and the humidity were in the range of 15-25°C and 70-75 % respectively.

The phytochemical investigation of hydro-alcoholic extract of whole plant of *Portulaca oleracea* carried out by standard procedures revealed the presence of alkaloids, flavanoids, glycosides, terpenes, saponins, carbohydrates, proteins, tannins, phenolics and steroids.

The results of the present study found that hydro-alcoholic extract of *Portulaca oleracea* reduced the glucose level in animals made diabetic with alloxan. Alloxan has been shown to induce free radical production and cause tissue injury. The pancreas is especially susceptible to the action of alloxan induced free radical damage. In the present investigation, hydroalcoholic extract of *Portulaca oleracea* demonstrated the significant anti-diabetic and hypolipidemic activity. The results from the present study also indicate that hydroalcoholic extract can reduce the levels of serum lipids. The antidiabetic effect of the hydro-alcoholic extract may be due to the enhanced secretion of insulin

from the beta cells of pancreas or may be due to increased tissue uptake of glucose by enhancement of insulin sensitivity.

The elevated plasma total cholesterol, triglycerides and LDL cholesterol are the major risk factors for the progression of cardiovascular diseases. Diabetic rats showed elevated plasma cholesterol, triglycerides and LDL cholesterol. Hydro-alcoholic extract in the dose of 200 mg/kg reduced the lipid profile along with the reduction in the blood glucose levels.

The literature reports reveal that flavonoids and tannins present in the plant extract known to possess antidiabetic and hypolipidemic activity. Since many antidiabetic drugs do not correct dyslipidemia, the observed hypolipidemic effects of the plant extract in diabetic rats makes *Portulaca oleracea* quite important in the management of diabetes. Since there is a strong well-established link between diabetes mellitus, dyslipidemia, obesity, hypertension and ischemic heart disease, effect of the plant extract on weight loss/gain needs to be explored on scientific base.

CONCLUSION

It has been concluded that the hydro-alcoholic extract of *Portulaca oleracea* has beneficial effects on blood glucose levels as well as improving hyperlipidemia and other metabolic aberrations. Further studies on pharmacological and biochemical investigations will clearly elucidate the mechanism of action and will help in projecting this plant as an therapeutic target in diabetics research.

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