



REVIEW ARTICLE

Assessment of Antidiabetic Activity of Dried Juice of *Andrographis Paniculata* Leaves in Alloxan induced Diabetic Rats

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ABSTRACT

Background: *Andrographis paniculata* (Acanthaceae) is an annual herb and the whole herb is traditionally reported to be used in treatment of diabetes mellitus.

Objective: To investigate the anti-diabetic potential of dried juice of *Andrographis paniculata* leaves in alloxan induced diabetic rats as the decoction of whole plant is used for the treatment of diabetes mellitus traditionally.

Method: The fresh leaves of *A. paniculata* herb were collected and grinded with distilled water into a fine slurry. The resulting juice was filtered and concentrated and used for the study. Three doses (50mg/kg, 100mg/kg and 200mg/kg) of dried juice of leaves were orally administered to the diabetic rats to evaluate blood glucose lowering effect. Metformin hydrochloride drug (120mg/kg) was used as a reference standard. The blood glucose level was measured at 0, 1, 3 and 5 hours after the treatment.

Results: After single administration of dried juice of leaves with 50 mg/kg dose there was significant reduction of blood glucose level at 3rd and 5th hour of administration and the % reduction was 24.8 and 37.51 respectively. Similarly 100mg/kg of extract dose showed significant reduction in blood glucose level at 1st, 3rd and 5th hour of administration with % reduction of 20.6, 35.2 and 48.28 respectively whereas 200 mg/kg dose caused significant reduction in blood glucose level after 1st, 3rd and 5th hour of intervals and the % reduction was of 21.48, 41.0 and 52.6 respectively.

Conclusion: All the three doses of dried juice (50, 100 and 200mg/kg) showed the maximum% reduction in blood glucose level after 5th hour of administration (37.51%, 48.28% and 52.6% respectively) as compared with diabetic control. This in vivo study shows that *A. paniculata* leaves has significant antidiabetic potential in a dose dependant manner and the results are comparable with the reference standard drug.

Keywords: Diabetes mellitus, Insulin, Alloxan, *Andrographis paniculata*.

Introduction

Diabetes mellitus is one of the most common disease affecting millions of people worldwide. It was known to Indian Ayurved as “Madhumeha” since 3000 years as a disease with some persons whose urine was sweet enough to attract insects and flies. It is a heterogeneous group of diseases characterized by a state of hyperglycaemia resulting from a diversity of etiologies, environment and genetic factors. It is a long term disease with variable clinical manifestations and leads to a number of complications like cardiovascular, renal, neuropathy, ocular and other intercurrent infections. (Irfan and Atiya, 2005)

Insulin is the main hormone which controls carbohydrate, protein and triglyceride metabolism and is secreted by the β cells of the pancreatic islets. The most striking acute effect of insulin is to lower blood glucose. Reduced or (absent) secretion of insulin causes diabetes mellitus.

Most patients can be classified clinically as having either type1 or type 2 diabetes and the vast majority of diabetic patients have type 2 diabetes. In recent years, predominantly due to lifestyle changes and the resulting surge in obesity, the developed nations have witnessed a tremendous increase in the occurrence of diabetes. (Brunton et al., 2006; Rang et al., 2016)

Diabetes mellitus has become a serious threat to human health and is often life threatening. In modern medicine various categories of synthetic drugs are available for its treatment but because of their inability to control all of the pathological aspects of diabetes, there is need to find out alternatives. (Marles, 1995; Chen, 2015)

Since time immemorial plant based medicines have been used by mankind to cure various diseases. As per the report of WHO, over 80% of the world population relies on the traditional systems of medicine, largely plant based to meet their primary health care and it has been globally recognized that medicinal plants play a significant role in providing health benefits to human beings. (Gupta and Ganguly, 2003)

In last few decades an increase in demand for plant based medicines and their eventual commercialization has brought about a major shift in their status and still at least 25% drugs derived from plants are included in the modern pharmacopoeia. The sector of medicinal plants has traditionally occupied an important position in the socio-culture, spiritual and medicinal arena of

rural tribal living of countries like India and China apart from other countries. (Agrawal et al., 2005)

Natural products are an abundant source for anti-diabetic drug discovery. By using proper scientific approach, the plant based anti-diabetic drugs especially used in folk medicine for treatment of diabetes may provide an alternative to synthetic drugs with low cost and less side effects. (Marles and Farnsworth, 1995; Chen et al, 2015)

Andrographis paniculata is an erect, annual, branched herb, up to 1 meter high, commonly known as Bhunimba in Sanskrit, Kalmegh/Kirata in Hindi and chiefly distributed in plains throughout India. The plant grows abundantly in moist, shady waste grounds and it is cultivated also. The stems are dark green in colour, quadrangular with longitudinal furrows and wings on the angles of the young parts. Leaves are dark green in colour, simple opposite, exstipulate, thin, membranous, up to 8 cm long, 2.5 cm broad, lanceolate, pinnate, upper surface glabrous, lower granular, margin entire to somewhat undulated, apex acute to acuminate, petiole short winged, flowers are small and fruit is capsule type.

The herb is a well known drug. It is official in I.P. and also employed in Ayurveda and Homeopathy systems of medicine. It is one of the constituents of Ayurvedic formulation “Sudarshan Churna”. (Nadkarni, 1982; Wealth of India; Quality Standards of Indian Medicinal Plants).

The herb is reported to contain various phtoconststiuents such as a bicyclic diterpenoid lactone-andrographolide, 14 deoxy, 11 oxo andrographolide, 14 deoxy andrographolide, neandrographolide, bisandrographolide, various oxygenated flavones, kalmeghin etc. (Rastogi and Mehrotra ,1993; Kokate et al., 2012)

The herb is reported to possess astringent, anodyne properties and is helpful in dysentery, cholera, diabetes, influenza, bronchitis, piles and gonorrhoea. Roots and leaves are used as stomachic, tonic, antipyretic, alterative, anthelmintic, febrifuge and cholagogue. (Nadkarni, 1982; Wealth of India)

Traditionally decoction of whole plant is given once a day for treatment of diabetes till cure. One spoon of mixture of leaves powder of *A. paniculata* along with powder of *Corallocarpus epigacus* tubers (2:1) is given with hot water twice a day for 7 days. (Irfan and Atiya, 2005)

Material and Methods

Collection and Authentication of Plant

Material

Andrographis paniculata leaves were collected from college campus of Smt. Kishoritai Bhojar college of Pharmacy, Kamptee, district Nagpur, Maharashtra, India and botanical identification of plant was done from Department of Botany, RTMNU, Nagpur whose Voucher specimen no. was 9532.

Preparation of Extract

A. paniculata leaves were thoroughly cleaned with distilled water. They were cut with scissor into small pieces and grinded into a fine slurry using distilled water in a grinder. The resulting juice was filtered and was concentrated under vacuum. A green solid mass was obtained.

Solution of dried juice was prepared by using normal saline for administration to experimental animals.

Preliminary Phytochemical Screening

The dried juice of leaves was subjected to preliminary phytochemical screening by means of standard chemical tests to detect presence of various primary and secondary plant metabolites. (Khandelwal, 2007)

Animals

Sprague Dawley rats (150-200 g) were used and kept at standard animal housing conditions of 12 hours light/dark cycle. Before experimentation, the animals were kept on fasting for 18 hours but allowed to have free access to water. The experimental protocol has been approved by the IAEC (Institutional Animal Ethical Committee).

Experimental Design

Alloxan induced Diabetes Model

The rats were randomly divided in 5 groups with six in each group. Diabetes was induced by single intraperitoneal injection of Alloxan monohydrate (150mg/kg). The rats with moderate diabetes were used in the experiment.

These groups were as follows.

GROUP-1: Untreated Diabetic control

GROUP-2: 120mg/kg reference standard Metformin HCl

GROUP-3: 50mg/kg dried juice

GROUP-4: 100mg/kg dried juice

GROUP 5: 200mg/kg dried juice

Blood sample were withdrawn after treatment at 0, 1, 3 and 5 hours from tail vein of rats and analyzed for blood glucose level using glucometer. (Vogel, 2002; Katsumata and Katsumata, 1990; Gupta, 2009)

Statistical Analysis

The results were expressed as mean \pm standard error of mean.

Statistical analysis of data was carried out by using one way analysis of variance (ANOVA) followed by Dennett's t-test. The results were considered to be statistically significant when p values <0.05 .

Results and Discussion

Preliminary phyto-constituents analysis of aqueous extract of *A. paniculata* leaves showed the presence of flavonoids, glycosides, tannins, saponins, alkaloids, and carbohydrates. (Table 1)

Table 1: Preliminary phytochemical screening of dried juice of *A. paniculata* leaves.

Sr. No.	Constituents	Results
1	Carbohydrates	+ve
2	Proteins	-ve
3	Alkaloids	+ve
4	Glycosides	+ve
5	Flavonoids	+ve
6	Tannins	+ve
7	Saponins	+ve

+ve indicates present, -ve indicates absent

The dried juice of leaves at 50 mg/kg showed a significant reduction of blood glucose level at 3rd and 5th hour of administration and the % reduction was 24.8 and 37.51 respectively. 100mg/kg of extract dose showed significant reduction in blood glucose level at 1st, 3rd and 5th hour of administration with % reduction of 20.6, 35.2 and 48.28 respectively whereas 200 mg/kg dose caused significant reduction in blood glucose level after 1st, 3rd and 5th hour of intervals and the % reduction was of 21.48, 41.0 and 52.6 respectively.

All the three doses of dried juice (50, 100 and 200mg/kg) showed the maximum% reduction in blood glucose level after 5th hour of administration (37.51%, 48.28% and 52.6% respectively) as compared with diabetic control. The reduction in blood glucose level was a dose dependent one and the results were comparable with that of the reference standard drug Metformin hydrochloride where the % reduction in blood glucose level was found to be 61.6 after 5th hour of administration. (Table 2 and Table 3).

Table 2: Anti diabetic effect of dried juice of *A. paniculata* leaves in alloxan induced diabetic rats.

Sr. No.	Group	Blood glucose level at hour (mg/dl)			
		0	1	3	5
1	Untreated diabetic control	321.8±29.6	339.1±19.19	354.83±17.5	409.0±19.4
2	Metformin HCl (120 mg/kg)	274.8 ±20.4	197.5±21.9***	132.6±9.0***	105.5±3.5***
3	Dried juice <i>A.paniculata</i> leaves (50 mg/kg)	273.0.± 20.4	296.6 ± 21.1	205.3 ±22.6***	170.6.±18.3***
4	Dried juice <i>A. paniculata</i> leaves (100 mg/kg)	248.6.±11.0	197.6.± 6.2***	161.3 ± 4.9***	128.6 ± 7.0***
5	Dried juice <i>A.paniculata</i> leaves (200 mg/kg)	242.6 ± 10.5	190.5 ± 4.5***	167.5±2.14***	115.0.±2.2***

Values are given as mean ±SEM (n=6), ***p<0.0001 compared with untreated diabetic saline control group.

Table 3: % Reduction in blood glucose level by different doses of dried juice of *A. paniculata* leaves and standard drug Metformin in diabetic rats

Treatment	Dose (Mg/Kg)	% Reduction of blood glucose		
		1h	3h	5h
Metformin HCl	120mg	28.13	51.75	61.61
Dried juice <i>A. paniculata</i>	50mg	--	24.8	37.51
Dried juice <i>A. paniculata</i>	100mg	20.6	35.2	48.28
Dried juice <i>A. paniculata</i>	200mg	21.48	31	52.6

In rural populations, the cases of diabetes mellitus are increasing tremendously throughout the world and there is an urgent need for the development of alternative strategies for diabetes therapy due to lack of ability of current therapies to control all the metabolic defects of the disease and also the great expense of modern therapy.

The primary and secondary metabolites of plant origin are widely distributed in nature and possess wide range of biological activities. The various plant metabolites like Glycans, steroids, alkaloids, saponins, polyphenols, flavonoids, terpenoids, coumarins etc. are reported to possess significant anti diabetic activity. (Marles and Farnsworth, 1995; Chen et al, 2015; Patel,2012)

The leaves of *A. paniculata* were found to contain flavonoids, saponins, tannins, glycosides, alkaloids so the blood glucose lowering effect of leaves can be attributed to the presence of these metabolites either singly or in combination.

Conclusion

From results, it is quite evident that dried juice *A. paniculata* leaves possess significant antidiabetic activity and it can be used for the management of diabetes. However, further studies are required to find the responsible active principle and its possible mechanism of action.

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Declaration of Interest

The authors report no conflict of interest.

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