Annals of Clinical Pharmacology & Toxicology

ISSN: 2642-9233

Research Article

Antidiabetic Potential of the Roots of the *Achyranthes Aspera* Linn.

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Abstract

Achyranthes Aspera linn is a vital source of drugs from the ancient time. It is known as Chirchita in hindi is an indigenous herb found in India, It has been used in almost all the traditional system of medicine, Ayurveda, Unani, and Siddha. The tribal, rural, and other people of our country commonly use this herb in various disorders. The leaves of the plant are reported to have antimicrobial property so mostly used in the treatment of skin and teeth disorder. The present study is aimed to evaluate the antidiabetic activity of the roots of this herb, which may help the researchers to set their minds for approaching the utility, efficacy and potency of herb. Only few researchers tried to evaluate antidiabetic potential of the leaves of *Achyranthes Aspera*. Therefore we selected this plant for the study of its antidiabetic potential. It was found that ethanol extract of the roots of *A.aspera* was active in STZ model in Sprague-dawley (SD) rats. Further to localize the activity in a fraction, the ethanolic extract was fractionated into four fractions and its chloroform fraction was the only most active fraction of the ethanol extract. After purification of the chloroform fraction, six compounds were purified. Out of six compounds two of them showed promising activity.

Keywords: Antidiabetic activity; *Achyranthes Aspera*; Roots; STZ rat model

Introduction

Achyrathes aspera Linn.(family; Amaranthaceae). In the country it is known by different names such as, Chirchita, Apamarga in Hindi. It has been used in all most all the traditional system of medicine. From the ancient time the tribal, rural and aboriginal people of our country used this herb in various disorders. It is an important plant of the traditional medicine in India. According to Ayurveda, it is bitter, pungent, heating, laxative, stomachic, carminative and useful for the treatment of vomiting, bronchitis, heart disease, piles, itching abdominal pains, ascites dyspepsia, dysentery, blood diseases [1,2].

The plant is an erect herb or under shrub up to 1 m high. Flowers are greenish white, in small dense axillary heads or spikes. Few triterpenoid saponins have been isolated from the plant having oleanolic acid as aglycone. Other constituents of the plant are ecdysterone, long chain alcohol, viz. 17-penta triacontanol, 27-cyclohexyl heptaeosan-7-ol, 16-hydroxyl 26-methyl heptacosan-2one and 36, 47-dihydroxy hen-pentacontan-4one. It also contains a water soluble base, betaine [3-9]. A number of workers have worked and reported various activities such as cardiovascular [10], effect on urinary tract [11], antibacterial [12], antifungal [12], antidiabetic [13], spasmolytic [14], antiasthmatic [15] antiallergic [16], diuretic [17] and many more are reported in the literature. The species has cooling, pungent, mild astringent, antiperiodic [17], digestive, purgative [17], laxative, abortifacient properties [18]. The paste of the root is given to stop bleeding after abortion [19] and to facilitate delivery and stimulate labor pain [19]. The decoction of the leaves is used in early stage of diarrhea and dysentery [20]. The paste of the leaves is externally applied over bites of poisonous insect, wasp, bees, burns and nephrotoxicity [21]. The seeds are used as an emetic, expectorant, brain tonic and is effective in biliousness and bleeding piles [22]. The ash of the plant is said to be effective in cough, chest pain and acidity [23]. It is also observed effective in abdominal tumor [24].

Citation: Lakshmi V, Srivastava AK. Antidiabetic Potential of the Roots of the Achyranthes Aspera Linn. Ann Clin Pharmacol Toxicol. 2019; 1(1): 1005.

Publisher Name: Medtext Publications LLC

Manuscript compiled: Feb 22nd, 2019

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Materials and Methods

Plant material

The roots of the *A.aspera* Linn. (2.0 Kg.) were collected from Lucknow, India and was authenticated by Botany Department of the Lucknow University.

Extraction fractionation and isolation of compounds

The air dried roots (1.0 Kg.) were powdered and percolated in 95% ethanol at room temperature for 24 hours, filtered and the process was repeated four times. All the extracts were mixed and filtered. Mixed ethanol extract was concentrated under reduced pressure below 50°C in a rotavapour to a green viscous mass, which was dried under high vacuum for 2 hours to remove the last traces of the solvent. Weight of the dried ethanolic extract 25.5 g. It was used for the screening of antidiabetic activity in STZ models in rats. Further the ethanol extract was fractionated into four fractions, hexane, chloroform, n-butanol soluble and, n-butanol insoluble fractions by maceration with the solvents in increasing order of their polarity (hexane→chloroform→n-butanol). The ethanol extract and chloroform fraction showed promising antidiabetic activity in STZ model. The chloroform extract on chromatography and rechromatography

ISSN: 2642-9233

Table 1: Antihyperglycemic effect of extract/fraction and pure compounds of *A.aspera* in streptozotocin induced diabetic rats.

S. No.	Compound	Dose(mg/kg)	% Antihyperglycemic activity	
			7 h	24 h
1	95% Aq. Ethanol extract	250	15.1**	26.6**
		500	20.6**	30.7**
2	Hexane soluble fraction	100	7.61	10.1
3	Chloroform soluble fraction	100	23.14**	28.4**
4	n-Butanol soluble fraction	100	10.8	12.7
5	Butanol insoluble fraction	100	4.88	3.53
6	Oleolenic acid	100	16.7**	18.7**
7	Ursolic acid	100	12.2*	15.9**
8	β-sitosterol	100	8.5	9.2
9	Triacontanol	100	6.2	8.6
10	Hexatriacontane	100	4.2	5.4
11	Palmitic acid	100	5.6	9.2
12	Metformin (Positive control)	100	26.4**	35.8***
13	Glybenclamide (Positive control)	100	23.6**	41.2***

Significance: ***p< 0.001 except marked with asterisk **p<0.01. *p<0.05

led to the isolation of 6 pure compounds. These compounds were characterized by physicochemical techniques (IR, NMR and Mass spectroscopy) and derivatization of suitable derivatives. Out of these 6 molecules tested only two showed promising results (Table 1).

Experimental animals

Sprague Dawley rats of either sex, weighing 180-200g were housed in elevated floor mesh cages to prevent coprophagy and were kept in environmentally controlled rooms (temperature 25 ± 2°C and 12 hours light and dark cycle rotation). All experimental protocols were approved by our Institutional Ethical Committee following the guidelines of CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals) which complies with International norms of INSA (Indian National Science Academy).

Streptozotocin induced antidiabetic rat model

Male Sprague Dawley strain albino rats of average body weight 140 ± 20 g having blood glucose profiles between 60 80 mg/dl were selected and were made diabetic by intraperitoneal administration of streptozotocin at a dose level of 45 mg/kg body weight. Streptozotocin (Sigma, USA) was dissolved in 100 mM citrate buffer (pH 4.5) and calculated amount of freshly prepared solution of streptozotocin was injected to overnight fasted rats. Blood glucose profile was again checked after 48 hours by glucostrips (Boehringer Mannheim) and animals showing blood glucose values between 150 to 450 mg/dl were included in the experiments and termed diabetic. The diabetic animals were divided into groups consisting of five animals in each group. Rats of experimental groups were administered suspension of the desired test sample prepared in 1% gum acacia at 100 mg/kg dose level. Animals of control group were given an equal amount of 1% gum acacia. Blood glucose profile of animals of all groups was again checked at 1, 2, 3, 4, 5, 6, 7 and 24 h post administration of test sample. Animals not found diabetic after 48 hours post treatment of the test sample were not considered and omitted from the calculations and termed as non-responders. Food but not water was withheld from the cages during the experimentation. Comparing the AUC of experimental animals as well as by comparing the data with results of standard drugs (Metformin and Glybenclamide) used for diabetic patients and the control groups determined the percent antihyperglycemic activity. Statistical comparison was made by Dunnett's test.

Conclusion

This paper explains evidence based-information regarding the antidiabetic activity of ethanol extract of the roots of *A. aspera* in STZ models. It was obvious from the result that, the ethanol extract of *A.aspera* played a beneficial role. Further the results shown in table-1 indicated that the chloroform fraction is the active fraction of the ethanol extract. On further purification of this fraction led to the isolation of 6 pure compounds(ursolic acid, oleonolic acid, sitosterol and triacontanol, hexatriacontane and Palmitic acid) out of which only ursolic and oleonolic acid shoed (table-1) potent anti diabetic effect in streptozotocin induced diabetic rats and could therefore be used as a remedy for the treatment of diabetes mellitus.

Acknowledgement

The author (VL) gratefully acknowledge the University Grant commission, Government of India for financial help in the form of Emeritus fellowship. Authors are also thankful to the head, biochemistry department, King George's Medical University, Lucknow, India for providing necessary research facilities to carry out this work.

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ISSN: 2642-9233

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