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Acute Diuretic Activity Of Withania Somnifera (L.) Dunal Leaves in Normal Rats



Avijeet Jain Dept. of Pharmacognosy, BRNSS- Contract Research Centre B.R.Nahata College of Pharmacy, Mandsaur 458001 - MP (INDIA)

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Lokesh Deb, Sunita Panchawat, Anurekha Jain, V.B.Gupta Dept. of Pharmacognosy, BRNSS-Contract Research Centre, B.R.Nahata College of Pharmacy, Mandsaur 458001 - MP (INDIA)

ABSTRACT

This work was undertaken to investigate diuretic activity of *Withania Somnifera*. Aqueous extract of leaves (100 mg/kg) was taken for screening of diuretic activity in albino rats after defatting and detoxification with petroleum ether and chloroform respectively. Frusemide was used as standard drug. The aqueous extract has shown significant diuretic activity which may be due to presence of polar compounds in *Withania Somnifera*. © 2006 Trade Science Inc. - INDIA

KEYWORDS

Diuretic; Withania Somnifer, Frusemide; Solanaceae; Acute toxicity.

INTRODUCTION

Withania somnifera (L.) (Dunal) (Solanaceae) is widely distributed in northwestern India, gujrat, madhya pradesh, and punjab plains and extends to the mountaineous regions of himachal pradesh and jammu^[1]. Traditionally leaves are used for treatment of tumors, inflammation, psoriasis, bronchitis, asthma, ulcers, scabies, insomnia etc.^[2]. Leaves are also used as hypnotic in alcoholism and as an anthelmintic^[3]. Leaves are reported to contain withanone, somnitol, glucose, and inorganic salts containing

chloride, sulfate, nitrate, sodium and potassium^[4]. Leaves are also reported to contain several chlorinated withanolides^[5]. Quantitative HPLC analysis of withanolides has already been reported^[6,7]. Previous reports show that leaves have hepotoprotective anti-inflammatory^[8], antitumour^[9] and antigeno toxicity activity^[10].

The diuretic activity of the root powder of Withania somnifera has been proved [11], but the diuretic activity of leaves of Withania somnifera was not well reported. Hence an effort has been made to get scientific proof of diuretic activity of aqueous

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extract of Withania somnifera leaves.

MATERIALS AND METHODS

Plant material

Fresh leaves of *Withania somnifera* were collected from rural area of Mandsaur district of M.P., India and identified by scientist of Govt. Horticulture College, Mandsaur M.P., India. A specimen (BRNCP/W/001/05) was deposited in Dept. of Pharmacognosy and phyochemistry, B.R.Nahata College of Pharmacy (BRNCP) Mandsaur, M.P., India for reference.

Preparation of extract

Shade dried powdered leaves were first defatted with pet. ether for 72 hrs. in soxhlet apparatus and then extracted with chloroform for detoxification and finally extracted with dist. water for 48 hrs. The extract was concentrated *in vacuum* under reduce pressure using rotary evaporator and dried.

Phytochemical studies

Aqueous extract was subjected for phytochemical studies which reveal the presence of alkaloids, glycosides, flavonoids, carbohydrates, saponins, tannis, proteins and free amino acids^[12].

Animals

Wistar albino rats (150-200gm.b.w.) and swiss albino mice (25-30 gm. b.w.) of either sex supplied by Central Animal House, BRNCP, was used. Animals were maintained under standard environmental conditions and had free access to standard rodent feed and water. The experiments on animals were conducted in accordance with the internationally accepted principles for laboratory animal use and the experimental protocols duly approved by Institutional Animal Ethical Committee (918/ac/05/CPCSEA).

Determination of acute toxicity (LD₅₀)

The acute toxicity for aqueous extract of *Withania* somnifera leaves was determined in wistar mice, maintained under standard conditions. The animals were fasted overnight prior to the experiment. Wistar mice either sex were divided into seven groups of six animals each. The control group received normal saline

(2ml/kg, p.o.). The other groups received 100, 200, 400, 800, 1000 and 2000 mg/kg of test extract, respectively. Immediately after dosing, the animals were observed continuously for the first 4 h for behavior. They were then kept under observation up to 14 days after extract administration to find out the mortality.

Assessment of diuretic activity

The effect of aqueous extract was evaluated for diuretic activity according to the method of Lipschitz [13,14]. Three groups of six rats, each weighing 150-180gm, were fasted and deprived of water for 18 hrs. prior to experiment. On the day of experiment, animals were given normal saline orally (25ml/kg body weight) in which the aqueous extract was dissolved. Control group received saline only. Test group received aqueous extract (suspended in water using 1% tween-20) at a dose of 100mg/kg body weight, p.o. standard group received frusemide (10mg/kg body weight, p.o.). The route of administration was per oral. Immediately after dosing, the rats were placed in metabolic cages (3 in each cage) specially designed to separate urine and faeces. Animals were kept at room temperature of 25+ 2°C throughout the experiment. The urine was collected in measuring cylinders up to 5 hrs. after dosing. During the experiment, no food or water was made available to animals. The total volume of urine collected was measured for each group (TABLE 1). The percentage increase in volume of urine was calculated by using formula

% Increase = Volume of control – Volume of test / Volume of control x 100

Statistical analysis

TABLE 1: Diuretic activity of aqueous extract of W. somnifera leaves

S. No.	Treatment	Dose	Total volume of urine (ml/kg) at 5 hrs.	Percentage Increase
1.	Control (Saline)	25ml/kg	7.400 ± 0.1581	
2.	Frusemide	10mg/kg	14.275 ± 0.4029***	92.905
3.	Aqueous extract	100mg/kg	12.075 ± 0.1548***	63.175

Data are given in Mean \pm Sem, ***P<0.001, (n=6).

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All data were expressed as mean ± SEM. Comparison between groups of parametric data was made using ANOVA test and Student's t-test which ever is applicable. P value of 0.01 or less was considered as significant.

RESULTS AND DISCUSSION

In acute toxicity study, aqueous extract of Withania somnifera leaves showed no mortality at the selected doses so 100mg/kg dose was selected for in-vivo experiments as sub-maximal dose.

The percentage increase of urine volume was significant 63% (12.075ml/kg) but it was less than the standard diuretic frusemide 92% (14.275ml/kg) and more than control (7.4ml/kg) (TABLE 1). In the present study, the diuretic effect observed does not exclude the possibility that changes in the diuresis may occur as a consequence of the presence of polar drug compounds^[15]. So, diuretic activity of *Withania somnifera* may be due to some polar phytochemical constituents present in the extract.

On the other hand, the absence of acute toxicity allows us to confirm the security also to take this plant, since at a dose even 20 times over used in this study, no sign of toxicity was shown.

CONCLUSION

The present results support the use of *Withania* somnifera leaves as a diuretic agent. More studies will be necessary to get more information about the potential diuretic value of leaves of *Withania somnifera* and in particular on the role of some plant component chemicals, and to evaluate the effects of long-term administration on diuretic activity.

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