

Evaluation of diuretic activity on the leaves of *Vigna unguiculata*

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Abstract

Vigna unguiculata is an annual herb, which is mainly used as diuretic, laxative, galactagogue, aphrodisiac and appetizer. The aim of this study is to investigate diuretic effects on the leaves of *Vigna unguiculata* in normal rats. The dried leaf powder was subjected to successive solvent extraction with petroleum ether, chloroform, ethyl acetate and ethanol. Among these ethanolic extract were administered orally at a dose of 200mg/kg and 400 mg/kg body weight by performing acute toxicity studies and investigated for diuretic activity in wistar rats using furosemide as standard. The extract was found to produce significant diuretic activity. The diuretic effect was evaluated by measuring the urine volume and levels of sodium, potassium and chloride ions. The results obtained revealed that the ethanolic extract showed significant diuretic activity at a dose of 400mg/kg body weight by increasing the total urine volume and levels of sodium, potassium and chloride in urine when compared to standard drug. The findings concluded that *Vigna unguiculata* leaves exhibit diuresis and these findings support the traditional use of *Vigna unguiculata* leaves as diuretic agents.

Keywords: *Vigna unguiculata*, ethanolic extract, acute toxicity study, diuretic activity

Introduction

Medicinal plants as a whole or their parts are being used in certain diseases [1]. The extracts of many plants used in traditional medicine contain curative agent that is used in many modern medicines [2] Traditional system of medicine are popular in developing countries, India has about 45,000 plant species & among them, several thousands have been claimed to possess medicinal properties [3] Herbal medicines are in great demand in developed as well as developing countries for primary health care because of their higher safety & lesser costs [4] Diuretics are the drugs that increase the rate of urination. There are several classes of diuretics & they increase the excretion of water from the body by acting through different mechanism. Drug induced diuresis is beneficial in many life threatening disease conditions such as Congestive Heart Failure, Renal failure, Hypertension, Cirrhosis, Nephritic syndrome [5]. Traditional system of medicine is rich in treating renal problems [6] Eg: *Mangifera indica*, *Allium sativum*, *Mimosa pudica*, *Lepidium sativum*, *Achyranthes aspera* etc., *Vigna unguiculata* is an annual herb, which is commonly known as Karamani/ Caramunnipayira in tamil. The traditional uses of *Vigna unguiculata* in which various parts such as whole plant is used as diuretics, Seeds are also used as Diuretic, boiled and eaten, considered good food and to destroy worms in the stomach and also used as laxative, appetizer, galactagogue, tonic, aphrodisiac, Pulse are also used as Diuretic and used to destroy worms in the stomach. Literature review also indicated that diuretic activity of this species has not been clinically evaluated so far. So the present study aimed to evaluate the diuretic activity of leaves of *Vigna unguiculata* in rats.

Materials and Methods

Collection

Collection of leaves of *Vigna unguiculata* was done in the June. Taxonomic identification of the plant was done by the

botanist Prof P. Jayaraman, Tambaram, Chennai. The leaves were shade dried for 15 days. After drying leaves were pulverized to a coarse powder.

Extraction

The powdered leaves were subjected to extraction by continuous hot percolation method with the use of solvents of increasing polarity. First leaves are defatted with petroleum ether. Then it is air dried and subjected to chloroform ethyl acetate and ethanol extraction. The extract was thereafter concentrated under vacuum and air dried.

Animals

Adult wistar rats of either sex weighing about 180-200 mg/Kg were used. The animals were randomly allocated into four groups each containing 6 animals and kept in cages and housed under standard conditions of temperature, humidity and dark light cycle (12h-12h)

Experimental Method

Acute Toxicity Study

Wistar rats of either sex weighing between 180-200 mg/kg were taken. The control group received normal saline (2 ml/kg, p.o.). The other groups received 100, 200, 300, 600, 800, 1000, 2000, 3000 and 4000 mg/kg of the test extract respectively, as well as, extract fractions up to 2000 mg/kg, in a similar manner. Immediately after dosing, the animals were observed continuously for the first 4 hours for any behavioural changes. Thereafter, they were then kept under observation up to 14 days after drug administration to find out the mortality if any. As there were no changes in mortality and behaviour. Hence the dose was safe for the diuretic activity.

Diuretic activity

In this method wistar rats of either sex weighing about 180-200mg/kg were taken deprived of food and water for 18 hours

prior to the experiment, and then the rats were randomly divided into four groups each containing six animals.

Group I: Control group in which only normal saline was given (25ml/kg)

Group II: Standard group in which Furosemide was given (10mg/kg)

Group III: Test group I in which 200mg/kg of ethanolic leaf extract of *Vigna unguiculata*

Group IV: Test group II in which 400mg/kg of ethanolic leaf extract of *Vigna unguiculata*.

Immediately after administration, the animals were placed in metabolic cages (2 per cage), specially designed to separate urine and faeces. The volume of urine collected was measured at the end of 5 h. During this period, no food and water was made available to animals. The parameters taken were total urine volume, concentration of Na⁺, K⁺ and Cl⁻ in the urine. Na⁺ and K⁺ concentrations were determined by flame

photometer and Cl⁻ concentration was estimated by titration with silver nitrate solution (N/50) using 3 drops of 5% potassium chromate solution as indicator.

The results were expressed as mean values ± S.E.M. (standard error of mean) for pairs of rats. Statistical comparison was carried out by analysis of variance (ANOVA).

Result and Discussion

In acute toxicity studies, however, there was no mortality at any of the tested doses till the end of 14 days of observation which was shown in table 1. And in diuretic studies, from the results it was found that ethanolic extract was found to produce significant increase in urine volume and the excretion of sodium, potassium and chloride ions at the higher dose tested (400 mg/kg p.o.) which was shown in table 2. Parameters of diuretic activity was shown in table 3. Urine volume is shown in figure 1.

Table 1: Acute toxicity studies

Observation	30 Mins	4 Hrs	24 Hrs	14 th Day
Body weight	No change	No change	No change	No change
Preterminal deaths	Absent	Normal	Normal	Normal
Cage side observation	Normal	Normal	Normal	Normal
Motor activity	Normal	Normal	Normal	Normal
Convulsions	Absent	Absent	Absent	Absent
Piloerection	Absent	Absent	Absent	Absent
Righting reflex	Present	Present	Present	Present
Lacrimation	Normal	Normal	Normal	Normal
Salivation	Normal	Normal	Normal	Normal
Respiration	Present	Present	Present	Present
Skin colour	Normal	Normal	Normal	Normal
Diarrhoea	Absent	Absent	Absent	Absent
Loss of corneal reflex	Normal	Normal	Normal	Normal
Loss of pinnal reflex	Normal	Normal	Normal	Normal
Grooming	Absent	Absent	Absent	Absent
Sedation	Normal	Normal	Normal	Normal
Excitation	Normal	Normal	Normal	Normal

Table 2: Concentration of Sodium, Potassium and Chloride ions

Treatment	Dose (Mg/Kg) (P.O)	Urine Volume (MI)	Electrolyte Levels		
			Na ⁺ (mmol/lit)	K ⁺ (mmol/lit)	Cl ⁻ (mmol/lit)
Group-I	Control-Saline (25mg/kg)	3±0.021	24±0.211	27.21±0.032	36.02±0.031
Group-II	Standard-Furosemide(20 mg/kg)	5.9±0.312	142.02±0.032	83.2±0.035	102±0.052
Group-III	200 mg/kg	3.4±0.220*	112±0.321*	70.00±0.402*	92±0.325*
Group-IV	400 mg/kg	4.5±0.351**	132±0.406**	74±0.621***	99±0.253**

ANOVA values are expressed as *P< 0.05, **P<0.001 when compared to normal.

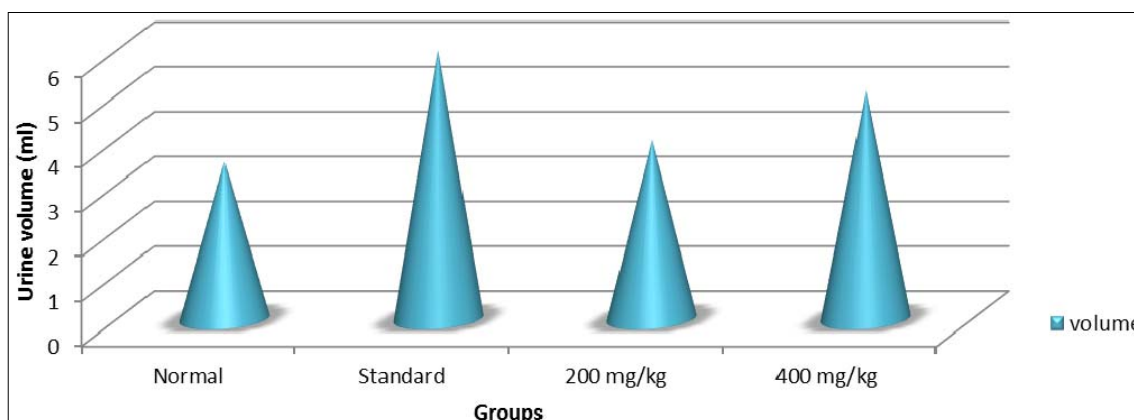


Fig 1: Urine volume

Table 3: Parameters of Diuretic activity

S. No.	Groups	Parameters		
		Na ⁺ / K ⁺	Na ⁺ + K ⁺	Cl ⁻ / Na ⁺ + K ⁺
1	Normal	0.88	51	0.40
2	Standard	1.71	225	0.55
3	200 mg/kg	1.6	182	0.50
4	400 mg/kg	1.69	206	0.52

Conclusion

From the above results it can be concluded that the ethanolic extract of *Vigna unguiculata* possess significant diuretic activity by increasing the urine volume and concentrations of sodium, potassium and chloride ions.

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