Available online on www.ijppr.com
International Journal of Pharmacognosy and Phytochemical Research 2023; 15(1); 10-14

eISSN: 0975-4873, pISSN: 2961-6069

Research Article

Diuretic Activity of an Aqueous Extract of *Cissampelos pareira* Roots in Albino Rats

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Received: 30th January 23; Revised: 19th February 23; Accepted: 26th February 23; Available Online: 25th March 23

ABSTRACT

Background: Diuretic chemicals are extremely beneficial in the treatment of congestive heart failure, nephritis, pregnancy toxaemia, premenstrual stress, and hypertension associated with oedema.

Aims: To investigate the diuretic effect of an aqueous extract of *Cissampelos pareira* roots in albino rats using the Lipschitz method.

Methods and Materials: Using metabolic cages, five groups of Albino rats were utilised to assess the diuretic efficacy of an aqueous extract of *Cissampelos pareira* roots. Group I received vehicle (normal saline of 5ml), Group II received Furosemide (10 mg/Kg, p.o), and Groups III, IV, and V received low (50 mg/kg), medium (100 mg/kg), and high (200 mg/kg) doses of aqueous extract of *Cissampelos pareira* roots, respectively. Immediately following the aqueous extract of *Cissampelos pareira* roots treatment, all rats were hydrated with saline (5 ml/kg, p.o.) and two animals were placed in each metabolic cage. Animals were not given food or water for 5 hours. At the end of 5 hours, the total volume of urine collected with each metabolic cage was measured. Various parameters such as total urine volume and ion concentrations such as sodium, potassium, and chloride in the urine were measured.

Results: When compared to the control group, the aqueous extract of *Cissampelos pareira* roots treated groups at different dose levels (50, 100, and 200 mg/kg) showed a significant increase in urine volume as well as a significant increase in the excretion of Sodium, Potassium, and Chloride ions in urine.

Conclusion: A single dosage of furosemide and an aqueous extract of *Cissampelos pareira* roots significantly (p0.05* & p0.01**) increased urine output while decreasing sodium, potassium, and chloride ion elimination. A 200 mg/kg aqueous extract of *Cissampelos pareira* roots demonstrated diuretic action significant to standard drug Furosemide.

Keywords: *Cissampelos pareira* roots, aqueous extract, hydrated rats, diuretic.

INTRODUCTION

Diuretic substances that increase water excretion have the potential to be beneficial in a wide range of illnesses, including those characterised by oedema, such as congestive heart failure, nephritis, pregnancy toxaemia, premenstrual tension, and hypertension[1]. The currently available diuretics, such as thiazides and loop diuretics, have a variety of side effects, including electrolyte imbalance and metabolic changes[2]. Most of diuretics are derived from medicinal plants, and a large number of medicinal plants mentioned in the Ayurvedic system of medicine like *Cissampelos pareira* etc are known to have diuretic properties.

Plant Information

Cissampelos pareira, a widely spreading, glabrous to soft pubescent, perennial climbing shrub distributed throughout India, is usually known as Padha, and additional synonyms include Padvel, Padvali, Aaknadi, Venievel, Poda, and Patha[3]. The leaves and roots are employed in the treatment of indolent ulcers (Kirtikar and Basu,) and diarrhoea (Amresh et al.,) in the Ayurvedic school of medicine. Because the plant is antiseptic, it is used to treat urinary tract infections (Dandiya and Chopra,). *Cissampelos pareira* juice is used to treat migraines, and the plant has a long history of use for muscle

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inflammation, snakebite, rheumatism, diarrhoea, dysentery, and menstruation difficulties.C*issampelos pariera* is commonly used in herbal therapy today as a diuretic, tonic, fever reducer, and pain reliever. Menstrual cramps, dysmenorrhea, heavy bleeding and uterine haemorrhages, fibroid tumours, pre and post-natal pain, colic, constipation, poor digestion, and dyspepsia are all common uses[3]. Several scientific studies have revealed that it has anticancer[4], anti-inflammatory[5], antidiarrheal[6], anti-hemorrhagic, antifertility[7], antioxidant, neuroprotective[8] antinociceptive, antiarthritic[9], cardiotonic[10], hepatoprotective[11], antioxidant, immunomodulatory[12], and antitrypanosomal[13] properties. The major constituents of roots of C.pareira include Pelosin, O-methylcurine, l-curine Cissamine, Cissampareine, Hyatin, Bebeerine, Cycleanine, Tetrandine and Berberine, Cissampeline, Cissampoline, Dicentrine, Insularine, Pareirine, Hyatinine, Pareirubrine A, Pareirubrine B, Pareitropone, Norimeluteine, Cissampeloflavone, D-Quercitol and Grandirubrine[13, 14]. C*issampelos pareira* roots are traditionally used as a diuretic but have not been properly studied as a diuretic agent. The primary goal of this study was to assess the diuretic effect of C*issampelos pareira* roots in hydrated (Modified Lipschitz test) albino rats.

METHODOLOGY

Extraction of roots:

Extract preparation involved thoroughly washing the roots under running water & shade dried. The dried collections of roots were powdered in grinder to a coarse powder. Used soxhletation to create an aqueous extract from the roots of C*issampelos pareira*. 200 grams of root powder were placed in the soxhlet device, the extraction procedure was run for 18 to 20 hours until a colourless solvent appeared in the side tube. By evaporating the solvents on a water surface, the collected extract was pooled & dried by concentrated under vaccum at 45ºc & stored in an airtight container for subsequent analysis.

Standard Drug: Furosemide 40 mg/ml (Sanofi, India Ltd) was used as a standard drug.

Experimental Animals:

Albino rats weighing 120-180 g of either sex were used in the study and were procured from the Central Animal House, Department of Pharmacology, Kakatiya Medical College**,** Warangal, Telangana, India. The experimental methodology was approved by the Institutional Animal Ethical Committee, and these animals were utilised to test the diuretic effect of an aqueous extract of *Cissampelos pareira* roots. The animals were kept under conventional husbandry settings for 15 days before the experiments with a temperature of 22+2°C. The experiment complied with laboratory's animal testing protocols and was authorised by the Institutional Animal Ethical Committee (IAEC).

Acute toxicity:

The acute toxicity[15,16] of an aqueous extract of *Cissampelos pareira* roots was tested using albino mice of either sex kept under conventional husbandry settings. The animals were fasted for overnight before to the experiment, and the extract was provided as a single dose, with mortality monitored for up to 48 hours (short term toxicity). The highest dose tested for LD50 (2000 mg/kg). To carry out this investigation, doses such as 1/20th, 1/10th, and 1/5th of the LD50 were chosen and considered as low, medium, and high doses, i.e. 50 mg/kg, 100 mg/kg, and 200 mg/kg, respectively.

Experiments[17,18]:

The Lipschitz Test was used to assess the diuretic activity of an aqueous extract of *Cissampelos pareira* roots in albino rats[19,20]. Male Albino rats were separated into five groups of six. The group I served as the normal control and received vehicle (normal saline 5ml/kg b.wt), the group II received furosemide (10 mg/kg, p.o.) in vehicle; groups III, IV, and V were treated with doses of 50mg/kg, 100mg/kg, and 200mg/kg of aqueous extract of *Cissampelos pareira* roots in vehicle and immediately following the extract treatment, all the rats were hydrated with saline (5 ml/kg) and placed in the metabolic cages i.e 2 animals per cage, which were specially designed to separate urine and faeces collection. At the conclusion of the six hours, the total volume of urine collected was measured. Animals were not provided with food or water during this time. Several factors, including the total volume of urine and the levels of sodium, potassium, and chloride in the urine were measured and estimated respectively.

Estimation of Urinary Electrolytes

In accordance with urine electrolytes (sodium, potassium, and chloride) were measured using the Ion Selective Electrode technique.

Analytical Statistics

The mean plus SEM (n=6) was used to express the experimental results. One-way ANOVA was used for statistical analysis, followed by the Dunnett's t-test.

RESULTS

To detect the phytoconstituents in the aqueous extract of *Cissampelos pareira* roots, qualitative phytochemical analyses were performed. The results showed the presence of sugars, alkaloids, sterols, phenolic compounds, tannins, flavonoids, and resins.

All of the rats in the acute toxicity investigation survived even 2 weeks later. As a result, it can be concluded that the extract is safe up to the highest dose level evaluated (2000 mg/kg). No

significant alterations in actions were observed throughout this study period.

Results from the evaluation of the diuretic activity of an aqueous extract of *Cissampelos pareira* roots are displayed in **[Table & Graph]** According to the findings, an aqueous extract of *Cissampelos pareira* roots significantly enhanced urine output and salt, potassium, and chloride excretion when compared to a control group. It was shown that the effects of the *Cissampelos Pareira* root aqueous extract were dosage dependent, meaning that among the three doses examined, the higher dose caused more effect.

The diuretic effect shown following treatment with an aqueous extract of *Cissampelos pareira* roots was found to be considerable in terms of urine output, salt, potassium, and chloride concentrations when compared to the conventional diuretic drug furosemide. Urine electrolyte concentration measurements showed that the aqueous-extract of *Cissampelos pareira* roots was successful in raising urine electrolyte concentrations for each of the three ions examined (Na+, K+, and Cl-).

For estimation of serum electrolytes blood was withdrawn from tail vein of the rat through a sterile syringe. Very gentle aspiration was done, in order to avoid vein collapse. Urine and blood electrolytes are estimated through flame photometer.

Table 1: Showing comparison of Urine Volume & Electrolyte Concentration

S. No.	Groups	Total Urine Vol (ml/kg b.wt /5 h)	Na^+ mmol/L	K^+ mmol/L	Cl^- mmol/L
1	Control (saline 5 ml/Kg b. wt)	12.54±0.02	118.03 + 1.18	52.09 + 1.21	80.59 + 1.42
2	Standard (Furosemide 10 mg/kg b.wt)	23.22±0.01***	193.05+2.10***	79.81+1.50***	125.60+1.87***
3	Aqueous extract of roots of *Cissampelos pareira* Low (50 mg/kg b.wt)	14.10±0.02	130.32+2.70	58.13+1.92	95.28 + 1.62
4	Aqueous extract of roots of *Cissampelos pareira* Medium (100 mg/kg b.wt)	16.14±0.02**	153.93+2.00**	76.93+2.67**	106.84+1.32**
5	Aqueous extract of roots of *Cissampelos pareira* High (200 mg/kg b.wt)	20.50±0.02***	175.43+2.22***	89.11+1.89***	120.59+2.03***

Values expressed as mean ± S.E.M., n=6, Significance at p<0.01**, p<0.001***, Compared with control group (One Way ANOVA followed by Dunnetts 't' test).

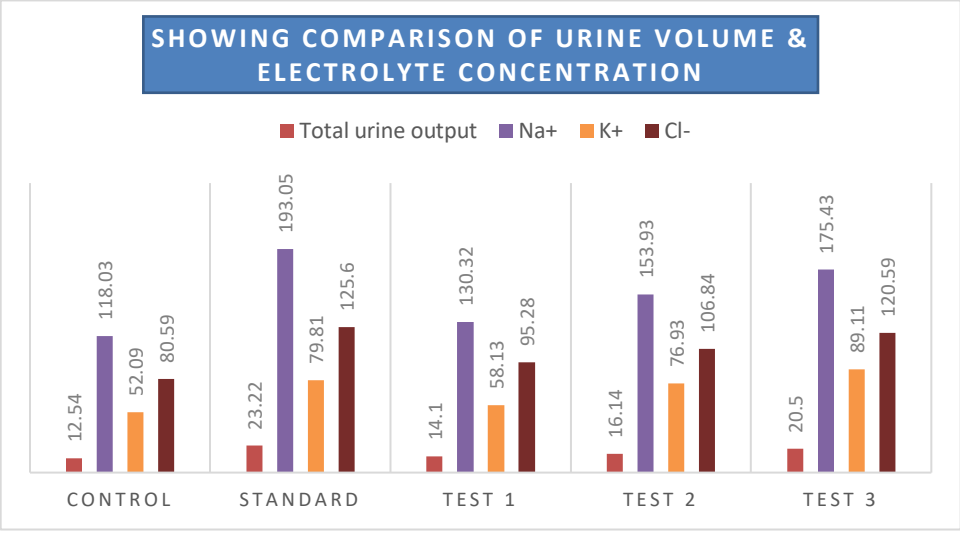

Figure 1: Graph showing comparison of Urine Volume & Electrolyte Concentration

DISCUSSION

Natural defences against disease are provided by medicinal plants and botanicals, which are also an important treatment for some illnesses. In addition to helping other antihypertensive medications work better, diuretics have proven to be incredibly helpful in the treatment of mild to moderate hypertension. Diuretics alleviate peripheral oedema and pulmonary congestion. These medications are helpful for treating acute left ventricular failure and CCF's orthopnea and paroxysmal nocturnal dyspnea[21]. They lessen plasma volume, which causes venous return to the heart to diminish. This lessens the pressure on the heart, the need for oxygen, the volume of plasma, and the oxygen demand. As a result, diuretics are crucial for patients with hypertension[22]. They are employed to cause forced diuresis (forced acidic and alkaline diuresis, respectively), in cases of aspirin and morphine poisoning.

Recurrent calculi can be treated with the help of diuretics. The results of this study showed that an aqueous extract of *Cissampelos pareira* roots considerably increased urine output and electrolyte excretion in a dose-dependent manner. Diuretic action using methonolic extract of *Cissampelos pareira* roots was previously reported by Suresh babu sayana et al.in 2014[3] & Hullatti et al. in 2011[2]. The diuretic efficacy of an alcohol extract of *Cissampelos pareira* was investigated in this work. The roots of *Cissampelos pareira* include flavanoids, alkaloids, carbohydrates, sterols, phenolic compounds, tannins, and resins, according to phytochemical investigations[23]. For this diuretic function, phytoconstituents like berberine[24] or pelosine have already been identified. Numerous plant extracts have been found to contain phytochemicals like flavonoids, saponins, organic acids, steroids, carbohydrates, tannins, phenolic compounds, terpenoids, alkaloids, glycosides, sterols, sesquiterpenes, and aminoacids[1,17]. The majority of these plant phytochemical compounds were found in Cissampelos pareira root aqueous extract. Therefore, it can be stated that the above phytoconstituents are responsible for the observed diuretic activity.

CONCLUSION

Results demonstrated that a single dose of normal furosemide (10 mg/kg) and an aqueous extract of *Cissampelos pareira* roots at doses of 50, 100, and 200 mg/kg each increased urinary output while also increasing the concentration of sodium, potassium, and chloride ions in urine. *Cissampelos pareira* root aqueous extract suggested a higher significant level of diuretic action that is comparable to that of conventional furosemide (10 mg/kg). The plant's diuretic properties are employed in traditional medicine. Our scientific study identified numerous phytoconstituents that were previously documented for this diuretic action in our aqueous extract of *Cissampelos pareira* roots. Thus, the folklore use of *Cissampelos pareira* roots for their diuretic activity is supported & justified by our study.

Conflict of interest: None

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