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EVALUATION OF DIURETIC ACTIVITY OF *DECALEPIS HAMILTONII* (WIGHT & ARN) ROOT EXTRACT.

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ABSTRACT

The present study is an attempt to investigate the diuretic effect of ethanolic root extract of *Decalepis hamiltonii* in wistar albino rats. The diuretic potential of ethanolic root extract was assessed in albino rats using in-vivo Lipschitz test model. The volumes of urine, urinary concentration of sodium and potassium ions were the parameters of the study. Frusemide was used as standard. The results indicate that ethanolic root extract at 400 mg/kg body weight shows a significant ($p < 0.05$) increase in the urine volume and electrolyte excretion ($p < 0.001$) when compared to control. Both (200, 400mg/kg) the extracts show significant diuretic activity. From the present study it may be concluded that the constituents present in ethanolic root extract of *Decalepis hamiltonii*(ERDH) may be responsible for diuretic activity.

KEYWORDS : *Decalepis hamiltonii*, Diuretic, furosemide.

INTRODUCTION

Man has been using herbs and plant products for its medicinal use since times immemorial. However, it is imperative that the traditional systems scientifically supported for their efficacy and safety. *Decalepis hamiltonii* belonging to family *Asclepiadaceae*, an endemic and endangered medicinal plant^[1]. The fresh roots of *D. hamiltonii* are available during monsoon in Southern parts of India and are generally dried and preserved for various food and pharmaceutical applications^[2, 3]. Roots of *D. hamiltonii* have traditionally been used

as demulcent, diaphoretic, diuretic and tonic. It is useful in the loss of appetite, skin diseases, diarrhoea, and nutritious disorders, as blood purifier^[4, 5], in the treatment of epilepsy and central nervous system disorders^[6]. Tuberous roots *D. hamiltonii* contains ellagic acid^[7], mainly volatile oil which contain 2-hydroxy-4-methoxy benzaldehyde, salicylaldehyde, benzaldehyde, methyl salicylate, benzyl alcohol, 2-phenylethyl alcohol, ethyl salicylate, p-anisaldehyde, vanillin^[8], ketone, resinol, sterols, saponins, tannins^[9], inositol^[10], fatty acids^[11], α -amyrin, β - amyrin

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acetate, and lupeol^[12]. Still no scientific and methodological investigation has so far been reported in literature regarding its action as diuretic agent. Therefore, the present investigation has designed to study the diuretic effect of root extract of *Decalepis hamiltonii*.

MATERIALS AND METHOD

Plant collection and Preparation of the Extracts

The roots of *D. hamiltonii* were collected from the herbal garden of Indian Institute of Horticultural Research, Bangalore. The roots were excised from the plants, washed in running tap water to remove adhered mud and sand then dry in shade and were grinded to get a coarse powder. The 200gms of powdered roots was soaked with petroleum ether for 2 days. At the end of second day the powder was taken out and it was dried. After drying it was packed in 1000ml soxhlet apparatus and extracted by using ethanol as solvent. The temperature was maintained at 55-65°C. After that extract was concentrated by distillation and solvent was recovered. The final solution was evaporated to dryness. The colour, consistency and yield (12.40% w/v) of ethanolic extract were noted.

Animals

Albino-Wister rats both sex (150-250g) were used with the approval of the institute animal ethics committee 1158/ac/07/CPCSEA. The animals were maintained in polypropylene cages of standard dimensions at a temperature of 28±1°C and standard 12 hour: 12 hour day / night rhythm. The animals fed with standard rodent pellet diet (Hindustan Lever Ltd) and water. Prior to the experiments, rats fed with standard food for 1 week in order to adapt to the laboratory conditions.

Acute oral toxicity study

The acute oral toxicity study has done according to OECD 423 guidelines. Administration of the extracts from 5 mg/kg up to the dose 5,000 mg/kg causes no considerable signs of toxicity in the tested animals. Dose of the lethal dose selected as the levels for examination of diuretic^[13].

Diuretic Activity

Male rats were (Wister albino strain) weighing 150 to 180gm maintained under standard condition of Available online on www.ijprd.com

temperature and humidity. The method of Lipchitz *et.al*,^[14, 15] employed for the evaluation of diuretic activity. The animals divide in to four groups of six rats in each and were fastened and deprived of food and water for 18 hr prior to the experiment. On the day of experiment, the group I animals served as control, received normal saline (25ml/kg,p.o), the group II animals received furosemide (100mg/kg,i.p)^[16], the group III animals received ethanolic root extract (200mg/kg,p.o) and group IV animals received ethanolic root extract (400mg/kg,p.o) respectively, in normal saline. Immediately after the administration the animals were kept in metabolic cages (three per cage) specially designed to separate urine and fecal matter and kept at room temperature of 25 ± 0.5° C throughout the experiment. The total volume of urine collected at the end of five hrs after dosing. During this period, no water and food made available to the animals. The parameters measured for individual rat were, body weight before and after test period, total concentration of Na⁺, K⁺ and Cl⁻ in the urine. The Na⁺ and K⁺ were measure by flame photometry^[17] and Cl⁻ concentration was estimated by titration with silver nitrate (N/50) using three drops of 5% potassium chromate solution as indicator^[18].

Statistical analysis

All the results expressed as mean ± standard error mean. The data analyzed statistically using ANOVA at a probability level of P < 0.001.

RESULTS AND DISCUSSION

Acute toxicity test

The LD₅₀ estimated to be greater than 2000 mg/kg. Hence, the biological dose fixed 200mg/kg and 400mg/kg for the extract.

Diuretic activity

Present study shows that the ethanolic roots extract possess good diuretic activity compared with standard drug furosemide. Here, the ethanolic roots extract increases the diuretic activity in a dose dependent manner. Na⁺/K⁺ ratio of 2.04 and 2.1 obtained for ethanolic root extract, respectively. The normal value for Na⁺/K⁺ ratio is reported to be 2.05 – 2.83. The concentration of aldosteron found to be dependent on Na⁺/K⁺ ratio.

If the Na^+ / K^+ ratio falls below the normal in plasma, the aldosterone secretion will be decrease and if the ratio rises above the normal value, the aldosterone secretion will be increase. Significant increase in total volume of urine and Na^+ , K^+ and Cl^-

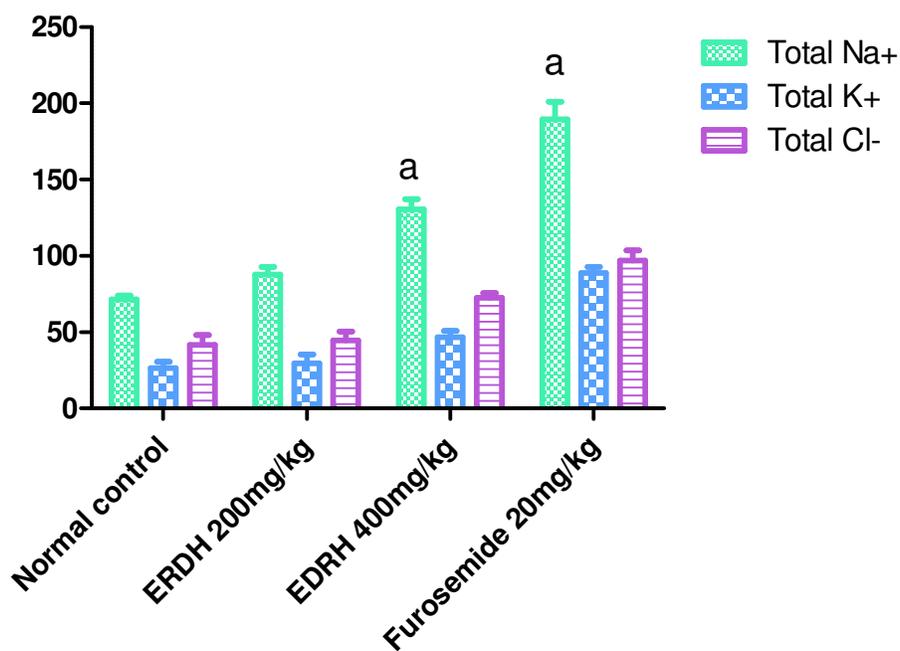
ion excretion observed in ethanolic root extract treated animals but it was less than the furosemide control. The results were shown in table 1 and figure 1.

Table: 1 Diuretic activity of ethanolic root extract of *Decalepis hamiltonii*.

Treatment	Dose	Total urine volume (ml/5hr)	Total Na^+ ($\mu\text{moles/kg}$)	Total K^+ ($\mu\text{moles/kg}$)	Total Cl^- ($\mu\text{moles/kg}$)	Na^+/K^+ ratio
Normal saline	25ml/ kg.	2.7ml	1985±16.02	904±9.82	629±16.01	2.195
Furosemide	100mg/kg	7.2ml	3496±6.2 ^{**}	1560±12.08 ^{**}	2688±18.54 ^{**}	2.241
ERDH	200mg/kg	4.2ml	2288±18.64 [*]	1118±10.11 [*]	2008±13.4 [*]	2.046
ERDH	400mg/kg	6.9.ml	3142±20.73 ^{**}	1494±8.43 ^{**}	2208±17.8 ^{**}	2.103

The value represents as mean ± S.E.M (n=6) animals of each group and was analyzed by ANOVA Tukey-Kramer multiple comparison test. In addition, the significance was conformed as ^{***} $P < 0.001$, ^{**} $p < 0.01$, ^{*} $p < 0.05$ by compared with control group.

Figure 1: Comparison of Total Na^+ , K^+ and Cl^- in Urine of various groups



The value represents as mean ± S.E.M (n=6) animals of each group and was analysed by ANOVA Tukey-Kramer multiple comparison test. ^a $P < 0.001$, compared with control group.

Diuretics relieve pulmonary congestion and peripheral edema. These agents are useful in reducing the syndrome of volume overload, decreases cardiac workload, oxygen demand and plasma volume, thus decreasing blood pressure^[19].

Thus, diuretics play an important role in hypertensive patients. In present study, we can demonstrate that ethanolic root extract of *Decalepis hamiltonii* may produce diuretic effect by increasing the excretion of sodium, Potassium and Chloride. The control of plasma sodium is

important in the regulation of blood volume and pressure; the control of plasma potassium is required to maintain proper function of cardiac and skeletal muscles^[20]. The regulation of Sodium, Potassium balance has intimately related to renal control of acid-base balance. The Potassium loss that occurs with many diuretics may lead to hypokalemia. For this reason, generally potassium-sparing diuretics recommended^[21]. In present study ethanolic root extract of *Decalepis hamiltonii* showed elevated levels of Potassium sparing capacity has investigated. Active principles such as flavonoids, saponins and terpenoids assumed responsible for diuretic activity.

In this study, diuretic action of ethanolic root extract of *Decalepis hamiltonii* was evaluated using furosemide as standard under controlled laboratory condition. As diuretic therapy may lead to be number of life threatening electrolytic disorder and toxicities, so safety profile studies are carried out by following a sub chronic administration of extract. This amplifies the heterogeneous array of diuretic curatives available for safe and effective treatment of edema and cardiovascular disease^[22].

CONCLUSION

The ethanolic root extract of *Decalepis hamiltonii* has showed the potent diuretic activity in Albino-Wister rats supporting the Ethanopharmacological use as diuretics. This effect may be exploring in the use of the plant in the management of some cardiovascular diseases.

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