Pharmacognosy Magazine
ISSN: 0973-1296

PHCOG MAG.
An official Publication of Phcog.Net

PHCOG MAG.: Research Article
Diuretic activity of whole plant extracts of *Cardiospermum halicacabum (linn)*

Chandra Prakash K.[1], Kuppast I.J.[2], Manjunath C.[3], Jawahar N.[1], Jubie S.[1]and Swapna B.[1]

[1] *Department of Pharmacology, S.A.C. College of Pharmacy, B.G. Nagara*
[2] *Department of Pharmacology, National College of Pharmacy, Shimoga*
[3] *Department of Pharmacology, Sharada Vilas College of Pharmacy, Mysore*

INTRODUCTION

Cardiospermum halicacabum (Linn), family Sapindaceae, is a deciduous, herbaceous climber, which is distributed through out the plains of India. The whole plant has been used for several centuries in the treatment of rheumatism, stiffness of the limbs, snake bite (1-3); its roots for nervous diseases, as a diaphoretic, diuretic, emetic, emmenagogue, laxative, refrigerant, stamachic and sudorific (4); its leaves and stalks are used in the treatment of diarrhoea, dysentery and headache and as a poultice for swellings (5). Phytochemical constituents such as flavone aglycones (6), triterpenoids, glycosides and a range of fatty acids and volatile esters (7-9) have been reported from the extracts of this plant. However the plant has not been experimentally tested for its diuretic property. Hence an effort has been made to investigate the same with whole plant extracts in experimental animal model.

MATERIALS AND METHOD

Drugs and Chemicals

Analytical grade petroleum ether and 95% ethanol S.D. Fine chemicals, Mumbai), glass distilled water and furosemide (Aventis, Mumbai) were used for the study.

Plant Extraction

The whole plant was collected from August to December 2005 and identified by Professor Srivastava, a botanist from LVD College, Raichur. A sample specimen was deposited, bearing voucher number C-2515. The shade-dried plant material was powdered. The coarse powder was subjected to successive extraction with petroleum ether and alcohol in soxhlet apparatus at 60-80ºC and the mark obtained after alcoholic extraction was macerated with water to obtain an aqueous extract.

Phytochemical Investigation

The petroleum ether (PeCh), alcohol (OHCh) and aqueous (AqCh) extracts of *C. halicacabum (Linn)* were subjected to preliminary, qualitative phytochemical investigation. The percentage yield for PeCh, OHCh and AqCh were 2.26, 2.50 and 4.80 respectively.

Experimental animals

Swiss albino mice (18-22 g) and Wistar albino rats (150-200 g) of either sex were acclimatized for 7 days under standard husbandary conditions, i.e. room temperature 35±1ºC, relative humidity 45-55% and light:dark cycle 12:12 h. The experimental protocols were approved by the Institutional Animal Ethical Committee (IAEC) of SAC College of Pharmacy, B.G. Nagara and Committee for the Purpose of Control and Prevention of Experiments on Animals (CPCSEA).

Acute toxicity Studies

The acute toxicity of PeCh, OHCh and AqCh was determined in female albino mice (18-22 g). After administration with different doses of these extracts, the mortality with each dose was noted at 48 hours (acute) and at 14 days (chronic). LD_{50} was calculated as per OECD guidelines 425 (10) using AOT 425 software.

Diuretic activity

The method described Lipschits et. Al (11). was employed for the assessment of diuretic activity. Health albino rats of either sex (160-200 g) were divided into five groups of six animals each. They were fasted 18 hours prior to the test, with free access to water. On the day of the experiment, animals were given 25ml/kg of body weight normal saline orally. Group I received vehicle (0.2 ml of 5% tween 80) and served as control group. Groups II, III, IV and V were treated with Standard drug (Furosemide 100mg/kg p.o.), PeCh (400mg/kg), OHCh (400mg/kg) and AqCh (400mg/kg) respectively. All drugs/vehicle were administered orally (p.o.).

Immediately after dosing, the rats were placed in the metabolic cages with special provision to collect faeces and urine. Animals were kept at room temperature of 35 ± 1^0C throughout the experiment.

The urine was collected in measuring cylinders upto a period of 24 hours. During this period no food or water was made available to the animals. The volume of total urine collected upto 24 hours was measured for both control and treated groups. The parameters taken to study were total urine volume, Na^+, K^+ and Cl^- concentration in urine. The sodium, potassium and chloride concentrations were measured colorimetrically (12-17) and the results were reported as mean ± SEM.

Statistical evaluation:

The groups were compared using one-way analysis of variance (ANOVA) followed by dunnett's test and $p<0.05$ was considered significant.

RESULTS:

It was found that the PeCh extract contained sterols, carbohydrates, tannins and triterpenes; the OHCh extract had sterols, saponins, carbohydrates, flavonoids and tannins; and the AqCh extract had sterols, saponins, carbohydrates, flavonoids and tannins.

In acute toxicity, there was no mortality recorded in all the groups, i.e. PeCh, OHCh and AqCh treated groups, up to the maximum dose of 2000 mg/kg. Hence $1/5^{th}$ of the maximum dose tested was selected for the diuretic activity.

All the three extracts have shown significant increase in urine volume, sodium ions excretion in rats but there is no significant alteration in pH and potassium ion excretion.

ANOVA studies have shown that there is a significant difference in diuretic activity among the groups. Dunnet "t" test indicates a significant difference in diuretic activity i.e. increased excretion of sodium ions, increase in urine volume in drug/extracts treated animals when compared with that in control animals.

The potency of diuretic activity follows the order standard > furosemide > aqueous > alcoholic > petroleum ether extracts.

DISCUSSION AND CONCLUSION:

Na^+/K^+ ratio of 1.76, 2.07 and 2.41 were obtained for Pet. Ether, alcohol and aqueous extracts respectively. The normal value for Na^+/K^+ ratio is reported to be 2.05-2.83 (18). The concentration of the aldosterone was found to be dependent on Na^+/K^+ ratio. If the Na^+/K^+ ratio falls below the normal in plasma the aldosterone secretion will be decreased and if the ratio rises the aldosterone secretion will be increased. Significant increase in Na^+ ion excretion was observed in Pet. Ehter, alcohol and aqueous extracts treated animals but it was less than the furosemide treated group. Flavonoids (21), saponins, volatile oils, sterols and triterpenes (22) are known to possess diuretic activity. The whole plant extracts (i.e. petroleum ether, alcohol and aqueous) of *Cardiospermum halicacabum L* contains flavonoids, saponins, sterols and triterpenes and these may contributed for the diuretic activity exhibited.

In the present study, the findings revealed that the tested extracts showed significant diuretic activity. Thus from the results of the current investigation it may be inferred that the Pet. Ether, alcohol and aqueous extracts of *Cardiospermum halicacabum Linn* possess diuretic activity. Further study regarding isolation and characterization of active principle responsible for diuretic activity at cellular level is needed.

Table-1 Diuretic activity of whole plant extract of Cardiosperrmum halicacabum (Linn)

Treatment	*Dose* Mg/kg	Volume of urine in ml	Na^+ mMoles/L	K^+ mMoles/L	Na^+/K^+	P^H
Control	25ml/kg	3.6±0.29	56.01±1.53	44.15±1.64	1.28±0.073	6.91±0.15
Standard	10	8.03±0.21**	102.38±1.87**	41.91±0.82	2.44±0.08	7.41±0.15
Pet ether Extract	400	4.48±0.46*	73.06±1.25**	41.73±1.42	1.76±0. 06	7.00±0.12
Alcoholic Extract	400	5.53±0.199**	85.22±1.54**	43.36±0.83	2.07±0.05	7.00±0.12
Aqueous Extract	400	6.86±0.18**	97.96±2.54**	41.01±2.00	2.41±0.0133	6.83±0.16

n=6, p< 0.05, p< 0.01***

The potency of diuretic activity follows the order standard > furosemide > aqueous > alcoholic > petroleum ether extracts.

References:

1. R.N. Chopra, S.L. Nayar and I.C. Chopra, ***Glossary of Indian Medicinal Plants*****,** (Council of Scientific and Industrial Research, New Delhi. 1986) pp.
2. ***Medicinal plants of Nepal***, (Dept. of Medicinal plants. Nepal 1993).
3. H.D. Neuwinger, ***African Traditional Medicine: A dictionary of Plant Use and Applications***. (Medpharm GmbH scientific Publishers, Stuttgart, Germany. 2000), pp. 1-300.
4. J.A. Duke and C.S. Ayensu, ***Medicinal plants of China.*** (Reference publications, Inc. 1985 ISBN 0-917256-20-4).
5. A. K. Nadkrani, ***Indian Materia Medica***, (Popular Book Depot, Bombay, 1976, reprint 2005), pp. 871-903.
6. M.M. Shabana, A.A. Genenah, S.M. El Zalabani Abou, R.G. El-Ela, M.F. Yousif Phytochemical investigation and insecticidal activity of Cardiospermum halicacabum L. cultivated in Egypt. ***Bulletin of the Faculty of Pharmacy*** (Cairo University), Vol. 28: 79-83 (1990).
7. I. Ahmed, M. Ahmad, A. Ahmad, Chemical investigation of the genus *Cardiospermum* of the family *Sapindaceae*. ***Science International***, Lahore, **5:** 67-69 (1993).
8. I. Ferrara, O. Schettino, D. Montesano, Triterpenoids from *Cardiospermum halicacabum L.* ***Phytotherapy Res***. **10:** S192-S194 (1996).
9. K. Srinivas, K.A. Choudhary, S.S. Rao, T. Satyanarayana, R.V. Rao, Krishna, Phytochemical investigation of *Cardiospermum halicacabum Linn.* ***Ind. J of Nat. Products*** **14:** 24-27 (1998).
10. OECD 2001-guidelines on acute oral toxicity (AOT) Environmental health and safety monograph series on testing and adjustment No. 425.
11. W.L. Lipschitz, Z. Hadidian, and A. Krepscar, Bioassay of Diuretics ***J Pharmacol. Exp. Ther.:*** 79-97 (1943).
12. R.F.L. IMaruna, ***Clin. Chem. Acta*****. 2:** 581 (1958).
13. P. Trinder, ***Analyst*** **76:** 596 (1951).
14. A.E. Terri, et. al. ***J Clin. Path.*** **29:** 86 (1958).
15. F.W. Sunderman, et. al. ***Am. J. Clin. Path.*** **29:** 95 (1959).
16. O. Schales, S.S. Schales, ***J. Biol. Chem.*** **140:** 879 (1941).
17. R.G. Schoenfeld, C.J. Lewellen, ***Clin. Chem.*** **10:** 553 (1964).
18. Anwar Massod, Siltanual-haq and Saxena S.K., Effect of some plant extracts on the larval hatching of methodology incognita chit wood. ***Indian Forester*** **111(10):** 841-845 (1985).
19. A. Doucet, A. Hus Chitharel, F. Morel, In vitro stimulation of Na-K ATPase in rat thick ascending limb by dexamethasone. ***Am. J. of Physiol.*** **(Renal Fluid Electrolyte Physiol. 20) 251:** F851-F857 (1986).
20. Doucet A, Katz A. I., Renal potassium adaptation: NA-K ATPase activity along the nephron after chronic potassium loading. ***Am. J of Physiol.*** **(Renal Fluid Electrolyte Physiol. 7) 238:** F380-F386 (1980).
21. Jianbo Xiao, Xinyu Jiang, Xiaqing Chen, Antibacterial, Anti-inflammatory and diuretic effect of flavonoids from *Marchantia convoluta*, ***Afr. J. Trad.*****,** CAM, 2, **3:** 244-252 (2005).
22. A. Tawfeq Al-Howriny, Studies on the pharmacological activities of an Ethanol extract of Balseenan (Commiphora opobalsamum), ***Pak. J Biol. Sci.*** **7 (11):** 1933-1936 (2004).