Research Article

Asian Journal of Biochemical and Pharmaceutical Research

Evaluation of Diuretic Activity of Fruit Extract of *Ziziphus jujuba* in Rats

Atul Kabra[1], Ruchika Garg[2] & Bharat Parashar[2]

[1] G.H.B Pharmacy College, Aniyad, Gujarat
[2] Departmrnt of Pharmacy Manav Bharti University, Solan, H.P.

Received: 19 January 2013; Revised: 04 February 2013; Accepted: 19 February. 2013

Abstract: *Ziziphus jujuba* is used in the traditional medicine as diuretic. In the present study, the diuretic activity of Petroleum ether, Chloroform, Alcohol extract of *Ziziphus jujuba* was studied and the activity was compared with furosimide as standard. The alcoholic extract exhibited significant diuretic activity as evidenced by increased total urine volume and the urine concentration of Na+, K+ and Cl-. The results thus support the *Ziziphus jujuba* use of as diuretic agent. These results clearly indicate that *Ziziphus jujuba* is effective against free radical mediated diseases.

Key Words: Diuretic Activity, Forosemide, Urine Volume, Flavanoids.

INTRODUCTION:

Herbs are used as medicine since time immemorial. Many of the natural products in plants of medicinal value offer us new sources of drugs which have been used effectively in traditional medicine. There is an increased consciousness regionally and globally in production and use of plants with healing property. Diuretics are the agents which cause an increase in the urinary output. These drugs are generally used in the treatment of hypertension, pulmonary and systemic edema [1].

DESCRIPTION OF PLANT [2]: Ziziphus jujuba is commonly known as Ber belonging to the family Rhamnaceae. Other names are:

Guajarati- Bordi, Hindi- Ber, Bengal-Kul, Kannad- Bogari.

Botanical classification [3]

Kingdom: Plantae Haeckel
Division: Magnoliophyta
Class: Magnoliopsida
Order: Rosales
Family: Rhamnaceae
Genus: Ziziphus
Species: Jujuba
Botanical name: *Ziziphus jujuba*

MATERIALS AND METHODS

Plant material: The fruit of *Z. Jujube* locally called as 'Ber' (in Hindi) were collected from the local market of Godhra in December 2011. The plant was identified by Dr. Himanshu A. Pandya, Associate Professor, University School of Science, Gujarat University, Ahmedabad, Gujarat. The reference no. of the specimen is GU/Bot/2012.

Preparation of plant extract: Fresh fruit of Ziziphus jujuba were collected. Seeds were separated from fruits and extract of fruit was prepared and extracted with petroleum ether, Chloroform and ethanol by maceration method and concentrated to get the residue. The yields of the extracts were 4.7, 3.5 and 2.7% respectively.

Animals: Male Wistar rats (175-200g) Male Balb/C mice (25-30g) were used for the experiments. They were housed in environmental conditions and fed with standard rodent diet and water *ad libitum.* All animal experiments conducted during the present study got prior permission from Institutional Animal Ethics Committee (IAEC) and followed the guidelines of IAEC (Ref: 1359/ac/10/CPCSEA).

Phytochemical analysis: Phytochemical analysis of the major phyto constituents of the plant extracts was undertaken using standard qualitative colour tests as described earlier [3,4].

Acute toxicity: Mice were divided into eight groups of six animals each. The control group received normal saline (2ml/kg, p.o.) The other groups received 50, 100, 200, 400, 800, 1000, 2000 and 4000mg/kg of the extracts, respectively. Immediately after dosing the animals were observed for their behaviour continuously for the first four hours. They were kept under observation up to 14 days after extract administration to find out the mortality and body weight [5].

.

Diuretic Activity: Male rats (wister albino strain) weighing 150 to 180gm were maintained under standard condition of temperature and humidity. The method of Lipschitz et al [6,7] was employed for the assessment of diuretic activity. The experimental protocols have been approved by the Institutional Animal Ethical Committee. Four groups of six rats in each and were fasted and deprived of water for eighteen hours prior to the experiment. The first group of animals serving as control, received normal saline(10ml/Kg, p.o.); the second group received furosemide (25mg/Kg, i.p.) in saline; the third, fourth, fifth groups received the Pet ether,

Chloroform, Alcohol extract at the doses of 200 mg/Kg, respectively, in normal saline. Immediately after administration the animals were placed in metabolic cages (2 per cage), specially designed to separate urine and feaces , kept at room temperature of 25± 0.5ºC through out the experiment. The urine was collected in measuring cylinders up to 3 hrs after dosing. During this period, no food or water was made available to animals. The parameters taken for individual rat were body weight before and after test period, total concentration of Na+, K+, and Cl- in the urine. Na+, K+ concentrations were measured by Flame photometry [8] and Cl- concentration was estimated by titration [9] with silver nitrate Solution (N/50)using three drop of 5% potassium chromate solution as

indicator. Furosemide sodium salt was given by stomach tube. Optimal dose activity relation was found to be 20mg/Kg of furosemide per kg body weight in series of supportive experiments. Results are reported as mean ± SD, the test of significance ($p<0.01$ and $p<0.05$) was stastically Table 3.

Statistical analysis: All the results are expressed as mean ± standard error. The data was analysed statistically using ANOVA followed by student 't' test [10] at a probability level of $P < 0.01$.

RESULTS:

Diuretic activity: The preliminary phytochemical analysis showed the presence of flavanoids, saponins, carbohydrates, terpenoids and alkaloids in all the extracts. The Alcohol extract 200mg/Kg p.o. showed significant increase in excretion of sodium, potassium and chloride ions in the urine in a dose dependent manner. The obtained effect was comparable to that of furosemide (25mg/Kg). Increase in urine output a sufficient index for assessing the diuretic effect through estimating the urinary concentration of Ion like Na+, K+, Cl- etc., may reveal in specific the Ion responsible for the diuretic activity. The results reveals that electrolyte excretions and diuretic activity of various extract of *Ziziphus jujuba* treatment possess significant diuretic activity at $P< 0.01$, but when compared to petroleum ether and Chloroform extract, the Hydro alcoholic extract possess more significant diuretic activity at $P<0.01$. Also all extract produced significant fall in potassium excretion compare to control.

DISCUSSION:

Diuretics relive 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 [11]. Thus, diuretics play an important role in hypertensive patients. In present study, we can demonstrate that ethanol, aqueous and chloroform extract 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 [12]. The regulation of Sodium, Potassium balance is also 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 are recommended [13]. In present study chloroform and alcohol extracts showed elevated levels of Potassium in urine, which may increase risk of hypokalemia and hence its potassium sparing capacity has to be investigated. Active principles such as flavonoids, saponins and terpenoids are known to be responsible for diuretic activity [14,15,16]. Results of present investigation showed that alcohol is most effective in increasing urinary electrolyte concentration of all the ions i.e Sodium, Potassium and Chloride followed by chloroform and pet ether extracts while other extracts did not show significant increase in urinary electrolyte concentration.

Table No. 1 Extractive Values

S. No.	Solvent	Colour of the extract	Percentage of yield
1	Petroleum ether	Dark Brown	4.7422
2	Chloroform	Dark Brown	3.5124
3	Alcohol	Dark Brown	2.4620

Table 2: Phytochemical Screening of *Ziziphus jujuba*

Extract	Pet.ether	Chloroform	Ethanol
Sterols	+	+	+
Terpenoids	–	–	–
Carbohydrates	–	+	+
Flavanoids	–	+	–
Proteins	–	–	+
Alkaloids	–	–	–
Glycosides	–	–	–
Tannins	–	–	+
Saponins	–	–	–
Phenolic compounds	–	–	+
Fixed oil and fats	–	+	+

(+) Presence of Constituents (-) Absence of Constituents

Table 3: Electrolyte excretion and diuretic activity of various extracts of *Ziziphus jujuba*

Group	Treatment	Dose	Urine Volume	Excretion Na+ Mg/lit	Excretion K+Mg/lit	Cl-Mg/lit	Na+/K+
G1	Normal control	100 mg/kg	8.0 ± 2.00	64.97± 2.33	53.76± 3.24	27.73±2.31	1.20
G2	Standard control	25 mg/kg	18.6 ±3.00	153.65± 4.97	22.09± 1.88	83.60±2.91	6.95
G3	Treatment control	200 mg/kg pet ether extract	8.9 ± 2.2	106.29± 5.32	38.25± 2.37	60.79 ±2.16	2.77
G4	Treatment control	200 mg/kg Chloroform extract	9.3± 2.6	120.30± 4.68	40 .40± 2.65	65.60±3.16	2.97
G5	Treatment control	200 mg/kg Hydroalcholic extract	12.8± 2.8	131.76± 3.62	45.47± 2.11	70.01±2.65	2.89

REFERENCES:

1. B.Danamma, K.Aruna kumari, B. Jayasimha goud & S. Nizamuddin Basha., *International Journal of Pharmacy and Biological Sciences*, 2011, **1(3),** 160.
2. KM. Nadkarni, *Indian Materia Medica*. 2nd ed. Bombay: Popular Prakashan., 1976,**vol 1**, 926.
3. GE Trease and WC Evans, *Pharmacognosy*, **13th Edn**. ELBS Publication , Delhi.,1989,171.
4. J.B Harbone ,*In; Phytochemical Methods A Guide to Morden Techniques of plant Analysis ,2nd* Edn.,Chapman and Hall, NewYork.,1984, 85.
5. U.K. Seth, N.K. Dadkar, U.G.Kamat., eds. *Selected Topics in Experimental Pharmacology*, 1st ed., Bombay, India, Kothari Book Depot, 1972, 124.
6. W.L. Lipschitz, Z. Haddian & A. Kerpscar., *J. Pharmacol. Exp. Ther.*, 1943, **79**, 97.
7. T. Murugesan, L. Manikandan, K.B.Suresh, M.Pal and B.P. Saha., *Indian J. Pharm. Sci*., 2000, **62(2),**150.
8. G.H. Jeffery, J. Bassett, J.Mendham, R.C.Denny, *Vogels Textbook of Quantitative Chemical Analysis*, 5th Edn. (Addison Westley Longman Ltd., England,1989) , 801.

9. A.H. Beckette, J.B.Stenlake, *Practical Pharmaceutical Chemistry*., Part I, Ist Edn., (CBS Publishers and Distributors ,New Delhi,1997) .,197.
10. P. Amritage., Eds., *In; Stastical Methods in Medical Research*, Blackwell Scientific Publications, London., 1971, 217.
11. R.D. Hoeland and M.J. Mycek., Lippincott's illustrated Reviews: Pharmacology, (Lippincott Willams and Wilkins, Philadelphia, 2000,157, 240.
12. A.C.Guyton & J.E.Hall., The body fluid compartments: extracellular and intracellular fluids; interstitial fluid and edema. In: Textbook of medical physiology, ninth edition. Singapore, PA: W.B. Saunders Company.,306,1998.
13. I.F. Sturat, Human Physiology, (Wm. C. Brown publishers, Dubuque, lowa Second edition)., 500, 508.
14. A. Chodera, K .Dabrowska, A. Sloderbach, L. Skrzypczak & J. Budzianowski., *Acta pol pharm.*, 1991; 48:35 37.
15. A.R.Sood, A. Bajpai & M. Digits., *Indian. J. Pharmacol*., 1985, **17 (3),** 178.
16. S.H Rizvi, A. Shoeb, R.S. Kapil & P. Satya., *Phytochemistry*, 1980, **19(11)**, 2409.

***Correspondence Author:** Ruchika Garg, Department of Pharmacy, Manav Bharti University, Solan, H.P.