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Research Article

COMPARATIVE DIURETIC ACTIVITY OF BADARASHMA BHASMA AND BADARASHMA PISHTI IN ALBINO RATS

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	ABSTRACT
<p>*Correspondence</p> <p>Ravi Mundugaru Research Officer, Department of Pharmacology and Toxicology, SDM Centre for Research in Ayurveda and Allied Sciences, Udupi, Karnataka, India</p> <p>DOI: 10.7897/2321-6328.02102</p> <p>Article Received on: 13/11/13 Accepted on: 21/12/13</p>	<p>Badarashma is an animal origin fossil. In Ayurveda it is used along with many herbal drugs for urinary disorders such as urinary calculi, urolithiasis. The present study was undertaken to compare the diuretic effect of two different preparations of Badarashma such as Bhasma and Pisti in albino rats to determine which of them would be good for therapeutic utilization. The Lipschitz <i>et.al</i> 1945 was used for the collection of data for diuretic action of the test preparations. Wistar albino rats were fasted for 18 h before starting experiment and suspensions of 2 preparations were administered at the dose of 45 mg/kg body weight. After treatment urine was collected for 24 h and diuretic activity was assessed by evaluation of the total volume of urine, sodium, and potassium and chloride concentration. The total urine volume of the rats treated with Badarashma Bhasma 45 mg/kg was found to be statistically significant when compared to control group and that of Badarashma pisti were found to be moderate when compared with the control. The excretion of cations such as sodium and anion chloride was found to be statistically non-significant with respect to control group but there was significant decrease in potassium excretion in Badarashma Bhasma. The observed diuretic effect was comparable to that of the standard drug furosemide but not the potassium sparing effect. The results obtained confirm presence of moderate diuretic activity in both the forms of Badarashma preparations. The bhasma preparation was found to be better diuretic than pisti.</p> <p>Keywords: Badarashma Bhasma, Badarashma Pishti, Diuretic activity.</p>

INTRODUCTION

Abnormality in fluid volume and electrolyte composition are common and important clinical disorders. Drugs that block specific transport functions of renal tubules are valuable clinical tools in the treatment of these disorders. A diuretic is an agent that increases urine volume, while natriuretic cause urine volume along with sodium excretion and usually called as diuretics¹⁻⁴. Most diuretic drugs have the adverse effect on quality of life including impotence, fatigue, and weakness^{5,6}. Although most of the diuretics proved to be very effective in promoting sodium excretion, all cause potassium loss and prompted the search for potassium sparing diuretic⁷. Hence search for a new diuretic agent that retains therapeutic efficacy and yet devoid of potassium loss is justified. Badarashma is an animal origin fossil obtained from Arabian countries which is used for treatment of various types of urinary diseases in different form. It is used effectively to treat the urolithiasis condition by Unani physicians from ancient days^{8,9}. It is claimed that,

Badarashma is having lithotriptic property; hence it will help to break the urinary calculi. When remnants of an animal or plant are exposed to the air or buried in dry earth, they generally decompose and pass off almost entirely as gases, but when buried under water or in damp earth, there will be preservation of these materials as fossil. Therefore, the species most likely to become fossilized are those living in water or marshes or in neighborhood of water or marshes. Badarashma is also called as Fossil encrinites. Here encrinites means fossilized stone. Badarashma is used in two different forms for the therapeutic purpose. One is in the form of Bhasma and other is Pishti form. Both these are considered to have Mootrala, mutrakrichrahara, ashmarihara properties^{10,11}. This may lead to confusion regarding selection form of the drug to be administered. The samples were assessed for diuretic activity along with this, electrolytes like sodium, potassium and chloride were also analyzed in the urine which are essential for maintenance of acid base balance through experimental study.

MATERIALS AND METHODS

Procurement and Preparation of test drugs

The raw materials for the preparation of trial drugs were collected from the S.D.M Pharmacy, Udupi, Karnataka, India. Badarashma Pisthi preparation was done according to Rasamrutham¹²⁻¹³. Bhasma was prepared according to the reference of Ayurvediya Rasashastra. The Badarashma Bhasma and Pisthi were prepared in the practical lab of Rasashastra and Bhaishajya Kalpana, S.D.M College of Ayurveda Udupi. The prepared drugs were mixed with 0.5 % gum acacia solution and administered to animals orally.

Dose selection and administration

Classical texts indicates, the dose of Badarashma Bhasma and Pisthi as 4- 8 ratti¹⁴. The dose of the experimental animals was calculated by extrapolating the human dose to animal dose as 45 mg/kg of rat based on the body surface area ratio by referring to the standard table of Paget and burners (1964)¹⁵. The study was carried out by administering the drugs orally with the help of feeding tube.

Animals

Wistar albino rats of either sex having a weight range of 160-240 g were used for the study. The animals were well housed in polypropylene cages under hygienic condition kept in 12 h light and dark cycle and maintained at 23- ± 2°C temperature. The animals were allowed have food (Pranav Agro Ltd' "Amrut" brand rat pellet) and water *ad libitum*. All the animals were acclimatized to laboratory conditions for a week before commitment of to the experiment. The institutional animal ethical committee approved all the experimental protocol (CPCEA/2011-RS01). The required numbers of Wistar albino rats for the study were obtained from the animal house attached to the SDM Centre for Research in Ayurveda and Allied Sciences and experiment was carried out in its Pharmacology laboratory.

Method

The method described by Lipschitz WL *et.al* (1945), Mukharji *et al.* (1996) and Murugesan *et al* (2000) was employed for the assessment of diuretic activity¹⁶. Healthy Wistar albino rats of either sex were divided into four groups of six animals each. The selected animals were fasted for 18 h prior to the experimentation with free access to water. On the day of experimentation rats were orally administered with normal saline 25 ml/kg body weight. Group I administered with vehicle and served as control group. Group II animals were administered with reference standard furosemide 20 mg/kg. Group III and IV administered with Badarashma Bhasma and Badarashma pisti in the dose of 45 mg/kg respectfully. After administration of group specific drugs all the rats were housed in individual metabolic cages in order to collect urine. Animals were kept at room temperature of 25 ± 5° C throughout the experiment. The urine was collected periodically in measuring cylinder up to a period of 24 hours. During these period animals were kept fasting without food and water. The parameters measured were total urine volume, sodium, potassium and chloride excreted in urine. The estimation of sodium, potassium and chloride was carried out with Flame photometer. The sum of sodium and chloride excretion as a measure of saluretic effect and the ratio of the concentration of sodium/potassium calculated for natriuretic activity and diuretic index of the end of 24 hours were calculated to assess the diuretic potential of the formulation.

$$\text{Diuretic index} = \frac{\text{urine volume in test group}}{\text{urine volume in the control group}}$$

Statistical analysis

The data obtained were analyzed by employing one way ANOVA followed by Dunnet's multiple t- tests as post hoc test. Graph pad Inst 3 was used for this purpose. A p value of less than 0.05 was considered to indicate statistically significant.

Table 1: Effect of Badarashma Bhasma and Pisti on excretory parameters

Treatment	Dose	Urinary excretion (ml)		Diuretic index
		4 th h	24h	
Normal saline	25 ml/kg	1.28 ± 0.25	2.75 ± 0.17	-
Furosemide	20 mg/kg	3.98 ± 0.81**	6.33 ± 0.79**	2.30
Badarashma Bhasma	45 mg/kg	2.27 ± 0.55	5.16 ± 0.55*	1.87
Badarashma Pisti	45 mg/kg	2.7 ± 0.4	4.41 ± 0.37	1.6

Values are expressed as MEAN ± SEM. N = 6. *P < 0.05 and **P < 0.01 when compared to normal saline group. ANOVA followed by Dunnet's multiple comparison test used for statistical analysis.

Table 2: Effect of Badarashma Bhasma on electrolyte excretion

Treatment	Dose	Electrolyte excretion		
		NA+ μMoles/L	K+ μMoles/L	Cl ⁻ μMoles/kg
Normal saline	25 ml/kg	130.56 ± 0.79	4.91 ± 0.26	107 ± 9.7
Furosemide	20 mg/kg	133.8 ± 0.87	4.97 ± 14	124 ± 4.7
Badarashma Bhasma	45 mg/kg	135.5 ± 1.61	4.13 ± 0.09**	120.8 ± 4.25
Badarashma Pisti	45 mg/kg	132.6 ± 2.28	4.08 ± 0.08**	133.26 ± 6.79

Values are expressed as MEAN ± SEM. N = 6. **P < 0.01 when compared to normal saline group. ANOVA followed by Dunnet's multiple comparison test used for statistical analysis.

RESULTS

In control group a gradual increase in urine output was observed during the observation period. Similar tendency was observed in both the test formulations and reference standard administered groups. In Badarashma Bhasma administered group, the urine output was found to increase during the observation period in time dependent manner. The urine output was found to be higher in comparison to the control group at the corresponding time intervals. The urine output recorded in this group at 4th and 24th h was found to be significantly high in comparison to the corresponding values of the control group. The 24 h urine output was found to be considerably increased both in Badarashma Bhasma and Badarashma Pishti administered groups in comparison to control group. However the Badarashma bhasma showed statistically significant increase in urine output in comparison to control group. The reference standard furosemide administered group showed consistence increase in the urine output at 4th and 24th hour reading and it is found to be statistically significant in comparison to control group. The urine sodium content was not found to be significantly altered by test drug and reference standard in comparison to control group. But urine potassium content exhibit significant decrease in these groups. Though comparatively higher level of chloride were observed in the urine collected from test drug and reference standard administered groups it was found to be statistically non-significant.

DISCUSSION

Urine output was assessed every hour up to 4 hours. Total urine output was measured after 24 hours without giving food and water. Urine output was comparatively more in Bhasma when compared to Pishti. But a constant increase in urine output was observed in Pishti group also. The results obtained in the present study clearly show presence of significant diuretic activity in Badarashma Bhasma and moderate and near significant activity with Badarashma Pishti, though both the formulation did not affect the electrolyte excretion in the urine. A good diuretic should excrete sodium along with water; however both the test drugs showed no significant effect on sodium excretion in urine in comparison to normal control group¹⁷. Surprisingly similar effect was observed with Furosemide also. The most frequent renal cause of potassium depletion is probably administration of diuretics without adequate potassium supplementation. All diuretics in common use except potassium sparing diuretics promote potassium excretion. Potassium excretion is increased during an osmotic diuresis. But both test formulation showed significant decrease potassium excretion it might indicate its potassium sparing action. The test drugs and reference standard though exhibited apparent increase in the chloride excretion the observed effect was found to be statistically non-significant. Based on the above activity profile- the probable site and mechanism can be suggested. Since no marked effect was observed on sodium excretion-interference with sodium re-absorption at loop of Henle and at other site does not seem to the site or mechanism of the diuretic activity observed. However, in the present study furosemide also did not enhance sodium excretion though it produced very good diuretic effect. This is a confounding

factor indicating the influence of the electrolyte balance maintenance. But for this the activity profile resembles that of potassium sparing diuretics or osmotic pressure which requires to be confirmed through further detailed study.

CONCLUSION

To conclude, it can be suggested that both the Bhasma and Pishti form of Badarashma possess moderate diuretic activity. Of the two, Bhasma has better diuretic potential than Pishti. Further detailed study is required to elucidate the exact mechanism of action the exact mechanism elucidation requires further detailed study.

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