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A Comparative Experimental Study to Evaluate *Mutrala* (diuretic) Activity of *Kusha* (*Imperata cylindrica* Beauv.) and *Darbha* (*Desmostachya bipinnata* Stapf.)

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Abstract

Objective: To evaluate the effect of *Kusha* (*Imperata cylindrica* Beauv.) and *Darbha* (*Desmostachya bipinnata* Stapf.) for *Mutrala* (diuretic) activity in experimental animals.

Methods: Three groups of six animals were used for experiment. Test drugs and vehicle were administered for one day. Diuretic activity was analyzed as per standard protocol.

Results: It was observed that from the two test formulations administered, *Desmostachya bipinnata* possessed moderate diuretic activity whereas *Imperata cylindrica* had only weak effect.

Conclusion: It can be concluded from the data generated during the study that *Desmostachya bipinnata* produces moderate diuretic effect which is much higher in magnitude in comparison to the weak diuretic effect observed with *Imperata cylindrica*. Also *Desmostachya bipinnata* has the potential to influence serum electrolyte content especially potassium and chloride content

Keywords

Mutral, Diuretic, Kusha, Darbha

Greentree Group

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INTRODUCTION

Kusha (*Imperata cylindrica* Beauv.) and *Darbha* (*Desmostachya bipinnata* Stapf.) are the constituents of the Trinapanchmoola[1] which is a well known diuretic drug in the Ayurvedic classics employed for various disease conditions like Shotha, Mootrakruchchhra, Ashmari etc. It is also has been included in Mootravirechaneeya Dashemani of Charak Samhita[2], the group of drugs which are useful diuretics for many pathological conditions. The individual action of both Kusha and Darbha as diuretics is also well documented in various Nighantus. As they are well known classical diuretic drugs it is considered worth to study them pharmacologically for assessment of diuretic activity to provide experimental basis to clinical findings and also to know probable mechanism of action.

MATERIALS AND METHODS

Procurement and preparation of test drug

The rhizomatous roots of *Kusha* (*Imperata cylindrica* Beauv.) and *Darbha* (*Desmostachya bipinnata* Stapf.) (Family - Poaceae)[3] were collected from the Killa Pardi region of Valsad district and Sasoi region of Jamnagar district and authenticated by Shri A.P.G. Pillai, (OSD), PGT-SFC cell I.P.G.T. & R.A., Gujarat Ayurved University. The rhizomatous roots were made into small pieces, shade dried, pulverized to fine powder (mesh number 80) and stored in airtight glass container for experimental purposes.

Animals

Wistar strain albino rats of either sex weighing 160-220g were used for the study. The animals were obtained from the animal house attached to the Institute. They were maintained on "Amrut" brand animal pellet feed of Pranav Agro Industries and plain tap water given *ad libitum*. Animals were exposed to natural day and night cycles with ideal laboratory condition in terms of ambient temperature (22 ± 2ºC) and humidity (50 – 60%). All the experiments were carried out between 8:00am to 1:00pm hours of the day. The experiments were carried out after obtaining permission from "Institutional Animal Ethics Committee"(IAEC/M.D.-03/07-08)

Dose fixation and route of administration

The dose selection was done on the basis of body surface area ratio using the table of Paget and Barnes (1969)[4]. Therapeutic human dose × Surface area ratio (Convertibility factor) for rat as required. Conversion of the dose obtained above to

dose in mg/kg/day by multiplying with suitable conversion factor based on the average weight of the animal. Human dose of both the drugs – 10000g/day[5]. Dose for Rats: 10000 mg × 0.018 = 180 mg / 200g rat = 900 mg/kg body weight of rats.

Preparation of test sample

Stock solutions of samples were prepared by dissolving 10mg of dried sample in 10ml of distilled water.

Route of administration

The drugs were administered orally with the help of gastric catheter of suitable size (catheter no. - 06) sleeved on to a disposable syringe nozzle.

Grouping

The selected animals were grouped into three groups of either sex and each group comprised of six animals.

(1) Group I – Water control

(2) Group II – *Imperata cylindrica* Beauv. root powder – 900mg/kg/day

(3) Group III – *Desmostachya bipinnata* Stapf. root powder – 900mg/kg/day

Statistical analysis:

The obtained data has been presented as mean ± standard error of the mean, difference between the groups, statistically determined by Student't' test for unpaired data to assess the statistical significance between the groups. $P<0.05$ is considered statistically significant.

Experimental model:

The diuretic activity was assessed following the standard method of Gillard *et al.*[6].

Diuretics are the compounds, which increase the quantity of urine. Normal urine output in rats is very small (1-2ml/rat per day). Hence to get the measurable quantity, the animals are first hydrated. The urine output is increased after administration of diuretics. Increases in volumes of urine is measured and compared with normal urine output. The selected animals were allotted to different groups randomly and allowed free access to water and food till the start of the experiment. The test drug and vehicles were administered to respective groups depending up on the body weight. After thirty minutes they were administered distilled water in the dose of 2ml/100g body weight to ensure uniform hydration. Then they were placed in individual metabolic cages with netted floor and urine was collected for a period of five hours in conical flasks placed below the polythene funnel of the metabolic cages. Volume of the urine collected was noted and a sample was taken for the estimation of sodium, potassium and chloride. Na^+ and K^+ were estimated by flame photometry and

chloride level by titration. And also different parameters like specific gravity and pH were tested by using Ames multiple kits. In addition, blood from supraorbital plexus was collected, serum was separated and sent to biochemical laboratory for estimation of sodium, potassium and chloride.

Table 1 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on urine volume in hydrated rats

Group	Dose (mg/kg)	Urine volume (ml/100g)	% Change
Control	Q.S	1.13 ± 0.33	-
I. cylindrica	900	1.32 ± 0.39	16.81 ↑
D. bipinnata	900	1.76 ± 0.08	55.75 ↑

Data: Mean ± SEM ↑- Increase

RESULTS AND OBSERVATIONS

Table 1 shows data of urine volume for diuretic activity of the plants in which both the test drug treated groups' show an apparent and statistically non-significant increase in the urine volume in comparison to normal control rats. The magnitude of increase is much higher in *D. bipinnata* administered group.

pH in hydrated rats

Group	Dose (mg/kg)	pH	% Change
Control	Q.S	7.75 ± 0.32	-
I. cylindrica	900	7.88 ± 0.24	-
D. bipinnata	900	7.63 ± 0.13	-

Data: Mean ± SEM ↑- Increase ↓ - Decrease

TABLE 3 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on specific gravity in hydrated rats

Group	Dose (mg/kg)	Specific gravity	% Change
Control	Q.S	1.008 ± 0.000	-
I. cylindrica	900	1.005 ± 0.003	-
D. bipinnata	900	1.004 ± 0.003	-

Data: Mean ± SEM ↓ - Decrease

Table 2 and Table 3 shows that administration of test drugs did not affect the urine pH and urine specific gravity to significant extent in comparison to control group.

Table 4 shows that an apparent and statistically non-significant increase in urinary sodium excretion is observed in *I. cylindrica* treated group where as in *D. bipinnata* treated group marginal decrease in urinary sodium excretion is observed.

Table 5 shows that an apparent and statistically non-significant decrease in

potassium excretion in both the test drug treated groups in comparison to control rats.

Table 4 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on urinary excretion of sodium in hydrated rats

Group	Dose (mg/kg)	Sodium (mEq/lit)	% Change
Control	Q.S	0.07 ± 0.022	-
I. cylindrica	900	0.09 ± 0.006	27.78 ↑
D. bipinnata	900	0.07 ± 0.007	04.17 ↓

Data: Mean ± SEM ↑- Increase ↓ - Decrease

Table 5 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on urinary excretion of potassium in hydrated rats

Group	Dose (mg/kg)	Potassium (mEq/lit)	% Change
Control	Q.S	0.52 ± 0.27	-
I. cylindrica	900	0.24 ± 0.07	53.41 ↓
D. bipinnata	900	0.14 ± 0.04	73.82 ↓

Data: Mean ± SEM ↓ - Decrease

Table 6 shows an apparent and statistically non-significant decrease in chloride excretion in both the test drug treated group in comparison to control rats.

Table 6 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on urinary excretion of chloride in hydrated rats

Group	Dose (mg/kg)	Chloride (mEq/lit)	% Change
Control	Q.S	0.17 ± 0.090	
I. cylindrica	900	0.04 ± 0.010	74.62 ↓
D. bipinnata	900	0.04 ± 0.008	76.20 ↓

Data: Mean ± SEM ↓ - Decrease

Table 7 shows that compared to normal control rats both test drugs increase the sodium level in serum marginally which is found to be statistically non-significant.

Table 7 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on sodium level in serum of hydrated rats

Group	Dose (mg/kg)	Serum sodium (mEq/lit)	% Change
Control	Q.S	129.00 ± 6.48	-
I. cylindrica	900	138.75 ± 1.84	07.56 ↑
D. bipinnata	900	139.25 ± 1.38	07.95 ↑

Data: Mean ± SEM ↑- Increase

Table 8 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on potassium level in serum of hydrated rats

Group	Dose (mg/kg)	Serum Potassium (mEq/lit)	% Change
Control	Q.S	6.20 ± 0.10	-
I. cylindrica	900	6.53 ± 0.46	05.25 ↑
D. bipinnata	900	5.93 ± 0.06 *	04.44 ↓

Data: Mean ± SEM ↑- Increase ↓ - Decrease *P<0.05

Table 8 shows that in comparison to normal control rats *I. cylindrica* treated group shows a marginal elevation in serum potassium where as *D. bipinnata* treated group shows an apparent and statistically significant decrease in serum potassium.

Table 9 indicates that both the test drug treated groups show an apparent increase in chloride level of serum in comparison to normal control rats, however only the increase observed in *I. cylindrica* treated group is found to be statistically significant.

Table 9 Effect of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. root powder on chloride level in serum of hydrated rats

Group	Dose (mg/kg)	Serum Chloride (mEq/lit)	% Change
Control	Q.S	094.75 ± 4.75	-
I. cylindrica	900	107.00 ± 1.68 *	12.93 ↑
D. bipinnata	900	105.75 ± 1.60	11.61 ↑

Data: Mean ± SEM ↑- Increase *P<0.05

DISCUSSION

Kusha (*Imperata cylindrica* Beauv.) and *Darbha* (*Desmostachya bipinnata* Stapf.) are the two important constituents of *Trinapanchmoola*, which is a potential diuretic combinational drug of Ayurvedic classics. The aim was to compare and assess the two test drugs for their diuretic activity. Diuretics are drugs that increase the rate of urine flow and excretion of electrolytes. The present study demonstrated that among the two drugs *Imperata cylindrica* did not show any effect on urine output in comparison to normal rats, whereas *Desmostachya bipinnata* increased the urine volume in non-significant manner. The increase was near significant- the reason for it not reaching statistically significant level is the variation in the data in the control group. Thus if considered in the light of this fact it can be suggested that *Desmostachya bipinnata* possesses moderate diuretic activity where as *Imperata cylindrica* has only a weak effect. *Desmostachya bipinnata* caused decrease in potassium excretion but the result was not statistically significant while *Imperata cylindrica* showed no significant effect on this parameter. Both the test drugs marginally enhanced the serum sodium levels in non-significant manner. This may be due to mild to moderate diuretic effect without the accompanying change in sodium excretion- thus this increase may be indicative of loss of body water without concurrent adjustment in the sodium level. *Desmostachya bipinnata* significantly decreased the serum potassium while *Imperata cylindrica* showed no significant

effect on this parameter. Several mechanisms have been identified that decrease the serum potassium level. Main among them are – decreased intake, loss in the gastrointestinal tract due to vomiting, diarrhea, discharge from the fistula, renal loss- as a result of excessive aldosterone, hydrocortisone, diabetic acidosis, magnesium depletion, renal tubular acidosis etc. The GI tract involvement can be ruled out since no diarrhea could be observed, diabetic acidosis can also be ruled out since the animals were normal, so also renal since no change in urinary potassium level could be observed hence the changes may be due to diet or changes in the magnesium level- these two parameters were not studied hence the assumption remains mainly presumptive in nature. In contrast to the changes in serum potassium level *Imperata cylindrica* significantly enhanced the serum chloride level where as *Desmostachya bipinnata* did not affect it. The exact reason is not known however, it can be suggested that it may be due to modulation of mineralocorticoid secretion from the adrenal cortex [Table 10].

CONCLUSION

The study showed that *Desmostachya bipinnata* Stapf. produces moderate diuretic effect which is much higher in magnitude in comparison to the weak diuretic effect observed with *Imperata cylindrica* Beauv. Further the data obtained reveal that *Desmostachya bipinnata* Stapf. has the potential to influence serum electrolyte content especially potassium and chloride content hence requires careful observation during clinical application.

Table 10 Consolidated statement of pharmacological profile of *Imperata cylindrica* Beauv. and *Desmostachya bipinnata* Stapf. for diuretic activity

Sr. no.	Parameters	*Imperata cylindrica*	*Desmostachya bipinnata*
1	Urine volume	NSE	NSI
2	pH	NSE	NSE
3	Specific gravity	NSD	NSD
4	Urine Na^+	NSE	NSE
5	Urine K^+	NSE	NSD
6	Urine Cl^-	NSD	NSD
7	Serum sodium	NSI	NSI
8	Serum potassium	NSE	**SD**
9	Serum chloride	**SI**	NSI

NSE – No significant effect; **NSI** – Non significant increase; **NSD –** Non significant decrease; **SD –** Significant decrease; **SI** – Significant increase

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