Nephro Protective, Diuretic and Anti inflammatory Evaluation of Monocot Grass *Kyllinga triceps* rottb

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**Abstract:** *Kyllinga triceps* rottb is commonly grown monocot grass of Gwalior Chambal region. In ayurvedic literatures it is known as musta. In various ethno botanical studies grass is reported to be diuretic, anti inflammatory,hepatoprotective and antidiabetic. In present study we have been evaluated kyllinga triceps rhizomes for its diuretic, nephroprotective and anti inflammatory potential.

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**I. Introduction**

Human beings are exposed intentionally and unintentionally to a various adverse chemicals which can harm the kidneys. As allopathic medicines, industrial chemicals, environmental pollutants can cause nephro toxicity. These nephrotoxics can produce a variety of kidney problems such as acute renal failure, chronic renal failure etc. Herbal diuretics is current demand of population for proper medication with out side effects. Diuretics are of immense importance with drugs given for treatment of blood pressure and heart problems. In the present study nephro toxicity is induced by administering gentamycin to rats. Plant is also evaluated for its diuretic and anti inflammatory potential and found to be potent diuretic and nephro protective agent. Present study introduce a new ignored herbal grass in the field of diuretic and nephro protective medication.

**II. Materials And Methods:**

**Plant Material**

Fresh rhizomes of *Kyllinga Triceps* Rottb. Were collected from Bhoora Khon area of Shivpuri District of Gwalior region, authenticated by Dr. (Smt) M.D. Gupta (Asst. Director) and Mr. N.K. Pandey (R.O.) National Research institute of Ayurveda and Siddha (CCRAS), Ministry for health and family welfare, Govt. of India, Amkho, Gwalior (M.P.)

**Ethical Aspects:**

The study was approved by the institutional ethical committee (protocol No. 891/Po/ac/05/CPCSEA).

**III. Pharmacological studies**

**Acute oral toxicy:**

Acute oral toxicity was performed by using OECD guide lines-423, fixed dose procedure (FDP). Five wistar albino rats of either sex having weight 175-200gm were used for the study. Fixed dose levels of 50, 100, 200, 500, 1000 mg/kg were given initially to allow identification of a dose producing evident toxicity for the ethanolic extracts of *Kyllinga triceps* rottb. LD 50 of the alcoholic extract of *Kyllinga triceps* rottb. Was done as per OECD guideline. The alcoholic extract falls under class 4 (LD50>2000mg/kg). The animals did not show any signs of toxicity and behavioral changes.

**Nephro protective Activity:**

**Experimental Animals:**

Male wistar albino rats 4-6 weeks, 175-200gm were used for the nephro protective evaluation of the ethanolic extract of rhizomes of plant *Kyllinga triceps*. the animals were maintained in well ventilated room temperature with natural day night cycle in the polypropylene cages. They were fed with balanced rodent pellet diet and tap water throughout the experiment animals were housed for 8 days, prior to the experiment to acclimatize the laboratory environment.
The rats were randomly divided into 3 groups each group contains 6 animals. 
Group I - It is served as normal group and 5 ml of distilled water is administered daily for 8 days. 
Group II- It is served as control group and gentamicin (100mg/kg/days) in given daily for 8 days through intraperitoneal route. 
Group III- It is served as test group and is given with ethanolic extract of kyllinga triceps rottb (400 mg/kg/P.O.) daily for 8 days concomitantly these rats are administered with gentamicin (100mg/kg/day) through intraperitoneal route 

After dosing on the 8th day, individual rats were placed in separate metabolic cages for 24 hrsto determine urine creatinine, urea, and uric acid content. Blood sample were collected via retroorbital puncture at the end of 24h, the serum was rapidly separated and process for determination of serum creatinine, serum urea and blood urea.

**TABLE-1** Nephro protective effect of ethanolic extract of rhizomes of plant kyllinga triceps on serum parameters.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Serum creatinine (mg/100 ml)</th>
<th>Serum uric acid (mg/100 ml)</th>
<th>Blood urea (mg/100 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group – I (Normal)</td>
<td>0.70 ± 0.01</td>
<td>0.30±0.02</td>
<td>23.40 ± 0.96</td>
</tr>
<tr>
<td>Group –II Gentamicin 100 mg/kg/day I.P</td>
<td>1.24 ± 0.05**</td>
<td>0.40 ± 0.02*</td>
<td>78.11 ± 0.92***</td>
</tr>
<tr>
<td>Group –III (Ethanolic extract 400 mg/kg/ P.O. with gentamicin 100 mg/kg/day, I.P)</td>
<td>0.89±0.02*</td>
<td>0.35±0.02*</td>
<td>30.03±0.2**</td>
</tr>
</tbody>
</table>

Values are expressed as MEAN ± SEM, one ways ANOVA followed by Dunnets”t test, note: n=6 in each group (* P value < 0.05, **p value < 0.01, *** P value < 0.001)

**Diuretic activity:** 
**Study protocol:** 
- Animal used : Albino rats 175-200 gms, 6 animals each group 
- Standard drug : Furosemide (20mg/kg) 
- Test drug : Ethanolic extracts of plantkyllinga triceps rhizome (100, 200 mg/kg)

**Experimental procedure:** 
The method of lipschizet.al was employed for the assessment of diuretic activity. Group of 6 albino rats, each weighing 175-200 gms were fasted and deprived of water for 18 hours prior to the experiments. On the day of the experiment all the animals were given normal saline orally 25 ml/kg body weight.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control</td>
</tr>
<tr>
<td>II</td>
<td>Ethanolic extract (100 mg/kg)</td>
</tr>
<tr>
<td>III</td>
<td>Ethanolic Extract (200mg/kg)</td>
</tr>
<tr>
<td>IV</td>
<td>Furosemide (20mg/kg)</td>
</tr>
</tbody>
</table>
Immediately after dosing, the animals were placed in metabolic cages specially designed to separate urine and feces and kept at room temperature of 25 ± 0.5°C. The urine was collected in measuring cylinder up to 5 hours after dosing. During this period no water or food was made available to the animals. The total volume of urine collected was measured for the control and treated groups. The parameters taken for each individual rat were body weight total urine volume, urine concentrations of Na⁺, K⁺, Cl⁻. Na⁺ and K⁺ concentrations were measured by flame photometry and Cl⁻ concentrations were estimated titrimetrically.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose</th>
<th>Urine Na⁺ ME q/Lt</th>
<th>Urine K⁺ ME q/Lt</th>
<th>Urine Cl⁻ ME q/Lt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Normal saline)</td>
<td>25 ml/kg</td>
<td>2.52 ± 0.09</td>
<td>96.2 ± 2.3</td>
<td>98.7 ± 0.94</td>
</tr>
<tr>
<td>Ethanolic extract</td>
<td>100 mg/kg</td>
<td>2.72 ± 0.14</td>
<td>120.2 ± 8.3</td>
<td>85.1 ± 0.28</td>
</tr>
<tr>
<td>Ethanol extract</td>
<td>200 mg/kg</td>
<td>3.25 ± 0.25*</td>
<td>125.6 ± 9.7*</td>
<td>87.7 ± 0.35*</td>
</tr>
<tr>
<td>Furosemide</td>
<td>20 mg/kg</td>
<td>3.55 ± 0.09*</td>
<td>138.8 ± 0.19*</td>
<td>94.6 ± 1.49*</td>
</tr>
</tbody>
</table>

* P > 0.01 Vs control by student „t” test.

Statistical Analysis:
The experimental results were expressed as the means± standard error of mean and the statistical significance was evaluated by using students „t” test.

Anti inflammatory Activity Study
Protocol:
Instrument used : Plethysmograph
Animal used : Albino rats 175-200 gms, 6 animals in each in four groups
Standard solution : Indomethacin (10mg/kg)
Control Solvent : Carrageenan 0.5 ml/kg
Test solution : Ethanolic extract 100 mg/kg and 200 mg/kg

Experimental Procedure:
Albino rats of either sex weighing 175-200 gms were divided into four group of six animals each. The dosage of the drugs administered to the different groups as follows.

Group- I : Control
Group- II : Ethanolic extract (100mg/kg)
Group- III : Ethanolic extract (200mg/kg)
Group- IV : Indomethacin (10mg/kg)

All the drugs were administered orally
After one hour of the administration of the drugs, dose 0.1 ml of 1% w/v carrageenan solution in normal saline was infected into the sub plantar tissue of the left hind paw of the rat and right hind paw serves as the control. The volume of the mercury displaced in the plethysmograph as measured at the end of 0 min, 120, min, 240 min, the % increase in paw edema of the treated group was compared with what of the control and the inhibitory effect of the drugs as studied. The relative potency of the drugs under investigations as calculated based upon the percentage inhibition of the inflammation.

Percentage inhibition:
Control (% increase in paw Volume in 3rd hour) - Test (% increase in paw Volume in 3rd hour) ×100

Control (% increase in paw volume in 3rd hour)
Nephro Protective, Diuretic and Anti inflammatory Evaluation of Monocot Grass Kyllinga triceps rottb

TABLE 3 Anti-inflammatory activity of ethanolic rhizome Extract of kyllinga triceps rottb on carageenan induced hind paw edema in rats

<table>
<thead>
<tr>
<th>Treatment (Dose)</th>
<th>% increase in paw volume (mean)</th>
<th>± (S.E.M)</th>
<th>% inhibition in paw volume</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (0.5 ml/kg)</td>
<td>28.73±</td>
<td>86.79±7.1</td>
<td>99.30± 3.8</td>
</tr>
<tr>
<td>Ethanolic extract (100mg/kg)</td>
<td>23.7±</td>
<td>63.9±5.3</td>
<td>68.7±4.9</td>
</tr>
<tr>
<td>Ethanolic extract (200mg/kg)</td>
<td>29.7±</td>
<td>49.3±2.8</td>
<td>59.3±2.3</td>
</tr>
<tr>
<td>Indomethacin (10 mg/kg)</td>
<td>27.8±</td>
<td>35.8±1.8</td>
<td>41.7±2.2</td>
</tr>
</tbody>
</table>

*P< 0.001 Vs control by student „t“ test.

Statistical Analysis
The experimental results were expressed as the mean ± standard error of mean (SEM) and the statistical significance was evaluated by using student „t“ test. The p-values of less than 0.001 imply significance.

IV. Results And Discussion:
The method of lipschitz et. al was employed for the assessment of diuretic activity ethanolic extract 200 mg/kg shows significant diuretic activity near to standard (Furosemide 20 mg/kg). Ethanolic extract shows better results. The ethanolic rhizome extract (200mg/kg) showed significant activity (p<0.001) at 2nd 3rd hours when compared to standard but it has been found that the dose of 200 mg/kg ethanolic rhizome extract showed good activity as compare to dose of 100 mg/kg ethanolic rhizome extract.
Nephrotoxicity has been induced by administering gentamicin (100 mg/kg) daily for 8 days. This has been reduced by giving ethanolic extract of the plant kyllinga triceps rottb. (400mg/kg) daily for 8 days.
Nephrotoxicity is identified by estimating the bio makers like serum creatinine blood urea and uric acid. Ethanolic extract of the rhizomes of plant kyllinga triceps rottb. Shows significant effect of the nephro protection due to its anti oxidant effect ethanolic extract has reduced the increased creatinine levels when compared to control group. Normal serum creatinine levels are 0.70 ± 0.01 that of gentamicin treated group is ± 0.05 and of plant extract treated group is 0.89 ± 0.02, levels of blood urea are 23.4 ± 0.96, 78.11 ± 0.92, 30.03 ± 0.2 in each group respectively.

V. Acknowledgment
Authors are thankful to Botany, Chemistry, Zoology and Pharmaceutical Sciences, Departments of Jiwaji University, Gwalior

Reference: