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A STUDY ON PRELIMINARY PHYTOCHEMICAL AND DIURETIC ACTIVITY OF BARK OF THESPESIA POPULNEA

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ABSTRACT

Thespesia populnea is a reputed ever green tree belonging to the family malvaceae; commonly known as Indian tulip tree. The plant is distributed tropical regions and coastal forest in India. It is well known and all the parts are used in traditional system of medicine. The plant has been used as astringent, antibacterial, hepatoprotective, haemostatic, anti-diarroheal and anti-inflammatory. The shade dried bark powder of Thespesia populnea was subjected to successive extraction using the solvents (Pet. ether, chloroform, alcohol and water). The diuretic potential of various extract of the barks was assessed in albino rats. The Volume of urine, urinary concentration of Na⁺, K⁺ and Cl⁻ ions were the parameters of the study. Fursemide (100 mg/kg) was used as standard. The extract (400 mg/kg) has shown significant increase in the volume of urine, urinary concentration of Na⁺, K⁺ and Cl⁻. From the present study it may be concluded that the extract possess polyphenolic compounds, carbohydrates, proteins and possess natriuretic and diuretic activities.

INTRODUCTION:

Thespesia populnea soland ex correa (family malvaceae) is a large tree found in the tropical regions and coastal forests in India and cultivated in the gardens. All the parts of the plant used in traditional system of medicine. The bark, leaves, flower and fruits are useful in cutaneous infection such as scabies, psoriasis, eczema, ringworm, and guinea worm. The decoction of the bark was commonly used for the treatment of skin and liver diseases. A compound oil of bark and capsules is useful in urethritits and gonorrhe¹.

The bark, root, fruits were used in dysentery, cholera and hemorrhoids. The fruits of the plant are used in Ayurveda for the control of diabetes². The barks and posses astringent, flowers hepatoprotective, antioxidant and antiinflammatory activities in rats^{3, 4, 5}. The leaves and bark of this tree are still used to produce oil for the treatment of fracture wounds and as an antiinflammatory poultice applied to ulcers and boils, as a folk medicine⁶. Gossypol was found to be the major component of Thespesia populnea producing fertility effects in rats 7, 8, 9 as well as in human beings¹⁰. Four naturally occurring quinones viz thespone, thespesone, mansonone-D, and mansonone-H have been extracted from heart wood of the plant¹¹.

The phytochemical study reveals the presence of carbohydrate, protein, tannins, phenol, flavonoids, terpenes, saponins and gums in the ethanolic extract of the bark¹².

In siddha system of medicine the plant bark has described to be used to reduce the swelling and in oedema of abdomin. So from this present study it may be conclude that the ethanolic extract posses a significant diuretic activity. The present study thus attempts to evaluate the bark and leaf of traditional medicinal plant *Thespesia populnea* (fam: malvaceae) which includes:

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- (a). To perform a pharmacognostical study, this is useful to evaluate the quality, purity and standard of the plant material. To isolate a possible new phytoconstituents, may verify their validity with their folklore claims.
- (b). To characterize the phytoconstituents and analyze by various instrumental methods.

MATERIALS AND METHODS:

Collection of plant material: The plant Thespesia populnea soland ex.correa belonging to family Malvaceae are widely distributed throughout tropical regions and coastal forest in India. The bark and leaf of *Thespesia populnea* soland ex.correa were collected from Selaiyur university campus in Chennai, Tamil Nadu in the month of January 2007. The species for the proposed study was identified and authenticated by Director, Plant Anatomy Research Centre, and Chennai. A voucher specimen (PARC/236/07) has deposited in the herbarium of the same department. The shade dried bark and leaves were cut it into small pieces and made into coarsely powdered using mechanical grinder and preserved in air tight container.

The fresh bark and leaf were used for macroscopical and microscopical study. The fresh leaf is utilized for quantitative microscopy. The powders of the plant material were used for physicochemical determination. The fresh barks were collected from our Selaiyur University campus in Chennai, India. The plant material was taxonomically identified and authenticated by Director, Plant Anatomy Research Centre, and Chennai. A voucher (PARC/236/07) has specimen deposited in the herbarium of the same department.

Phytochemical screening: The dried and powdered bark was subjected to preliminary phytochemical screening for qualitative detection of phytoconstituents. The dried and coarsely powdered bark (100 g) was extracted successively with aqeous extract, ethanol (90%), chloroform, and ethyl acetate in a soxhlet extractor by continuous hot percolation. Finally the marc was macerated with chloroform water.

Each time before extracting with the next solvent of higher polarity the powdered drug (marc) was dried in a hot air oven below 50°C for 10 minutes. Each extract was concentrated by distilling off solvent. which was recovered subsequently. The concentrated extracts were evaporated to dryness and the extracts obtained with each solvent were weighed. The crude extracts were analyzed for the presence of various phytoconstituents by following standard phytochemical tests¹³ and the results were reported (Table 1).

Table 1: Results of phytochemical screenings of successive extracts of bark of *Thespesia populnea*

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Constituent	Chloro- form extract	Ethyl acetate Extract	Ethanol extract	Aqueous extract
Alkaloids	-	-	-	-
Carbohydrates	-	-	+++	++
Glycosides	-	-	++	+
Steroids	-	-	-	-
Flavonoids	-	-	+	-
Saponins	-	-	++	-
Fixed oils and fats	-	-	-	-
Tannins	-	-	+++	++
Proteins and amino acids	-	-	-	+
Mucilage	-	-	-	+++

Pharmacological study:

Animals: Wistar rats (150-200gm) were purchased from King Institute, Chennai for experimental study. They were acclimated to animal house condition feed with commercial pellets Rats chon (Hindustan Lever Ltd, Bangalore, India) and had free access to water. The experimental protocol was approved by the IAEC (Institute Animal Ethical Committee) of CPCSEA (Committee for the Purpose of Control and Supervision of Experiment on Animal).

Acute toxicity studies: Acute oral toxicity study was performed as per OECD-423 guidelines (acute toxic class method). Wistar rats (n=6) of either sex selected by random sampling techniques were employed in this study. The animal were kept fasting for overnight providing only water. Then the bark extracts were administered orally at the dose of 2000 mg/kg by intragastric tube and observed for 2 days for the gross behavioral changes and mortality¹⁴.

Experimental protocol:

Group I – Administered with normal saline 10 ml/kg, p. o.

Group II – Administered with aqueous extract of bark 400 mg/kg, p. o.

Group III – Administered with ethanol extract of bark 400 mg/kg, p. o.

Group IV – Administered with chloroform extract of bark 400 mg/kg, p. o.

Group V – Administered with ethyl acetate extract of bark 400 mg/kg, p. o.

Group VI – Administered with standard drug furosemide 100 mg/kg, p. o.

Immediately after the respective treatments the animals were placed in metabolic cages and urine was collected in the measuring cylinder up to 5 hr. The volumes of urine, sodium, potassium and chloride ions were estimated in the urine for assessing diuretic activity.

Diuretic activity: Albino rats of either sex weighing 150 to 200 gm were divided in to

six groups of six animals each. The animals were fasted for 24 hrs and water was given ad libitum during fasting. On the day of experiment the animals of group I was administered with saline (25ml/kg p. o.) and this group served as control. Similarly the animals of group II, III and IV, V, VI were administered with aqueous, ethanol, choloform, ethyl acetate and test extracts of 400mg/kg orally (as a suspension of 0.5% CMC) and furosemide 100mg/kg (standard). **Immediately** after respective treatments the animals were placed in metabolic cages (3 animals in one metabolic cage) and urine was collected in the measuring cylinder up to 5 hrs. The volume of urine, Na, K and Cl were estimated in the urine for assessing diuretic activity (Table 2)^{15, 16, 17}.

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RESULT:

The phytochemical studies indicated the presence of carbohydrate, protein, tannins, phenol, flavonoids, terpenes, saponins and gums in the ethanolic and aqueous extract of the bark. Ethanolic extract has increased the volume of urine significantly at 400 mg/kg. The results of the present study are in conformity with the reports that the plant possesses flavonoids like β sistosterol, etc.

The Na⁺ and K⁺ ion excretion is significantly elevated. But Cl ion excretion was not elevated significantly. The results are indicating that the extract is potent natriuretic. However the natriuretic effect is sufficient to cause diuresis. The diuretic effect of the test extract was significantly lesser than that of frusemide 100 mg/kg (standard). However, the contribution of polyphenolic compounds to diuretic effect

cannot be ruled out. Further studies like isolation and characterization of diuretic principle from the barks of the plant is needed to confirm. From the study it may be concluded that the claim of the native practitioners that, the leaves possess diuretic effect, is justifiable.

CONCLUSION:

The aqueous, chloroform, ethanol, and ethyl acetate extracts were found to be active on the renal system in rodents. The data in the table allow the conclusion that the extracts were aquaretic. The values of urine volume were elevated. This is also valid for the reference substance, furosemide. However the action excretion is increased. Furosemide increased the sodium excretion two times, while the extract at the dose of 400 mg/kg by 1-2 fold. Potassium excretion level was significantly (P< 0.05) increased in comparison with water control. A very high increase for the chloride excretion was also observed. For the diuretic activity, the results clearly indicate that the extract of Thespesia populnea acted as an aquaretic in rats, but enhances considerably ion excretion almost to an extent similar to that produced by furosemide.

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Table 2: Diuretic activity of various successive bark extract of *Thespesia populnea*

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Group	Total urine	Total Na ⁺	Total K [†]	Total Cl ⁻
	(ml)	(mmol/kg)	(mmol/kg)	(mmol/kg)
-	4.02±0.02	420±25	1542±108	762±308
п	9±0.8*	1618±118*	1630±228 ns	1612±108*
Ш	8.88±1.2*	1157±212*	2024±142 [*]	1892±116 [*]
IV	C 22±0 48 ^{ns}	1004±100*	1894±168 [*]	1407±172*
IV	6.32±0.48	10041190	1894±108	148/I1/2
V	9.40±2.1	1796±160 T	1618±192 [*]	1516±168 [*]
VI	12.32±0.22*	2412±42*	1950±336 [*]	3078±218 [*]

The values are mean ±SD of 6 animals in each group.

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 $^{^{*}}$ P < 0.05, ns – non significant, Comparisons were made between group I vs. group II, III, IV, V and VI

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