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Phytochemical analysis of methanolic leaf extract of *Thespesia populnea*

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Abstract

Thespesia populnea is a herbal medicinal plant known for its therapeutic value in inflammatory disorders. The present study was carried out to know the qualitative phytochemical analysis of methanolic leaf extract of *Thespesia populnea*. The leaves of *Thespesia populnea* were collected from in and around Tirupati (Chittoor district), Andhra Pradesh. The plant was identified and authenticated by a taxonomist in the Department of Botany, College of science and arts, Sri Venkateshwara University, Tirupati. *T. populnea* methanolic leaf extract was prepared by using cold maceration method and subjected to analysis of carbohydrates, phenolic compounds, flavonoids, alkaloids, glycosides, saponins, steroids and proteins. In the present study, methanolic extract of *Thespesia populnea* leaves revealed the existence of various phytochemical constituents like carbohydrates, phenolic compounds, flavonoids, alkaloids, glycosides, saponins, steroids and proteins.

Keywords: *Thespesia populnea*, phytochemical, methanolic extract, leaves, medicinal use

Introduction

Medicinal plants possess spectrum of active principles and are useful as curatives in various human and animal diseases. The continuing use of herbs in medicine reveals the functional value and its necessity in the future. In modern medicine, the importance of medicinal plants is increasing. *Thespesia populnea* commonly called as Hibiscus populnea belongs to the Family: Malvaceae. *Thespesia populnea* is an evergreen tree. The Leaves are alternate, simple, with petioles of length 5-10cm long and shiny, oval or triangular in shape (Archana *et al.*, 2010) [2]. The flowers are hibiscus like single at upper leaf axils, corolla yellow with a red centre. The Fruits are Globose. It is a large tree found in the tropical regions and coastal forests in India and cultivated in the gardens (Parthasarathy *et al.*, 2009) [17]. All the parts of the plant used in traditional system of medicine. The bark, leaves, flowers and fruits are useful in cutaneous infection such as scabies, psoriasis, eczema, ringworm, and guinea worm (Chang *et al.*, 2002) [5]. The decoction of the bark is commonly used for the treatment of skin and liver diseases. A compound oil and capsules made from bark is useful in urethritis and gonorrhoea. The bark, root and fruits were used in dysentery, cholera and haemorrhoids (Chabra *et al.*, 1994) [4]. An ayurvedic preparation containing *Thespesia populnea*, namely "panchvalkala" possess free radicals. The bark and flowers possess astringent, hepatoprotective, antioxidant and anti-inflammatory activities (Shirwaikar *et al.*, 1995, Illavarasan *et al.*, 2003, Satyanarayana *et al.*, 2004 and Manivasudevan *et al.*, 2007) [20, 8, 19, 14]. The main chemical constituents are Kaempferol, Quercetin and its glycosides, herbacetin and its glucoside, populneol, populnin, populnetin, rutin, gossipetin, gossypol, lupeol, sesquiterpenoidal quinones viz; thespeson, thespone, mansonones C, D, E and F, amino acids and carbohydrates. Fruit juice is used on rheumatism sprains, scabies, swellings, insect bites and warts. Pulp of fresh fruits were applied for relief of migraine. Unripe fruit juice was used to cure piles. Decoction of bark was given to treat diarrhoea and arthritis. Root, fruit and leaf were used in psoriasis, scabies and other cutaneous diseases. Bark was used for the treatment of hemorrhoids and chronic dysentery. Leaf is used for an anti-inflammatory effect (Asima and Satyesh, 1994 and Jean, 1999) [3, 10]. The leaves are applied locally on swollen joints for their anti-inflammatory effects and also for skin diseases, hepatitis, jaundice, ulcers, wounds, psoriasis, scabies, urinary tract infections, diabetes, cholera, cough, asthma and gunvaworm infections. Gossypol was found to be the major component of *Thespesia populnea* flowers (Akila and Rani, 1993). Hence, the present study was carried out to know the qualitative phytochemical properties of methanolic extract of *Thespesia populnea*.

Materials and Methods

The leaves of *Thespesia populnea* were collected from in and around Tirupati (Chittoor district), Andhra Pradesh. The plant was identified and authenticated by a taxonomist in the Department of Botany, College of Science and Arts, Sri Venkateshwara University, Hyderabad.

Preparation of methanolic leaf extract of *Thespesia populnea*

T. populnea methanolic leaf extract was prepared by using cold maceration method. The leaves of *T. populnea* were shade dried and ground to a coarse powder. About 100 g of leaf powder was soaked in 500 ml of 95% methanol (v/v) for 72 h with intermittent mixing using a glass rod and then filtered through muslin cloth followed by Whatman No. 1 filter paper. The filtrate was concentrated by rotary evaporator and then air dried. Extract was weighed and the percentage yield was calculated with reference to the air-dried material.

Qualitative Phytochemical examination of the extract

The extract obtained was subjected to preliminary phytochemical screening for the detection of various plant constituents by following standard procedures (Evans, 2002) [6].

Test for carbohydrates

Molisch's test: About 300 mg of extract was mixed with 4ml distilled water and filtered. To 2 ml of this filtrate, 2-3 drops of alpha naphthalene solution in alcohol was added, shaken for 2 min and 1 ml of concentrated sulphuric acid was added slowly from the sides of the test tube. Then it was observed for the presence of purple ring at the junction.

Fehling's test: Fehling's A and Fehling's B solutions, each 1ml were mixed and added to 2 ml of extract, heated in boiling water bath for 10 min, appearance of yellow and then brick red precipitate indicated the presence of reducing sugars.

Test for Phenolic compounds

Ferric chloride test: Small quantity of the extract was mixed with water and treated with dilute ferric chloride (5%) and observed for the presence of blue colour.

Lead acetate test: Small quantity of extract was mixed with water and treated with 3 ml of lead acetate solution. The occurrence of white precipitate indicated the presence of phenols.

Test for flavonoids

Shinoda test: To 2 to 3ml of diluted extract, a piece of magnesium ribbon and 1ml of concentrated HCl was added. A pink or red coloration of the solution depicted the presence of flavonoids.

Test for alkaloids

To 10 g of dry extract, 20 ml of dilute hydrochloric acid was added, shaken well and filtered. The following tests were performed using the filtrate.

Hager's test: To 3 ml of filtrate, 1 ml of Hager's reagent (saturated picric acid solution) was added and observed for the presence of yellow precipitate.

Wagner's test: To 3 ml of filtrate, 1ml of Wagner's reagent

(iodine in potassium iodide) was added. The appearance of reddish brown precipitate indicates the presence of alkaloids.

Mayer's test: To 3 ml of the filtrates, 1ml of Mayer's reagent (potassium mercuric iodide) was added. The appearance of white precipitate indicates the presence of alkaloids.

Tests for Glycosides

Legal's test: Dissolved the extract (0.1g) in pyridine (2ml), added equal volume of freshly prepared sodium nitroprusside solution (2ml) and made alkaline with Sodium hydroxide solution. Pink to red colour solution shows the presence of glycosides.

Test for Saponins

Foam test: 1ml of extract was diluted with 20ml of distilled water and shaken in a graduated cylinder for a few minutes. A 1cm layer of foam formation indicates the presence of Saponins.

Test for steroid / terpenoid

Liebermann-Burchard's test: To 1ml of extract, 1ml of chloroform, 2 to 3 ml of acetic anhydride and 1 to 2 drops of concentrated Sulphuric acid were added. Dark green coloration of the solution indicates the presence of steroids and dark pink or red coloration of the solution indicates the presence of terpenoids.

Test for Proteins

Biuret test

To 0.5 mg of extract equal volume of 40% NaOH solution and two drops of 1 % copper sulphate solution was added. The appearance of violet colour indicates the presence of protein.

Ninhydrin test

About 0.5 mg of extract was taken and two drops of freshly prepared 0.2% Ninhydrin reagent was added and heated. The appearance of pink or purple colour indicates the presence of proteins, amino acids or peptides.

Results and Discussion

The methanolic extract of *T. populnea* was estimated for qualitative phytochemical analysis and is presented in Table 1 and Plate 1. In the present study, the methanolic extract of *Thespesia populnea* leaves revealed the existence of various phytochemical constituents like carbohydrates, phenolic compounds, flavonoids, alkaloids, glycosides, saponins, steroids and proteins. Similar results were reported by Krishnamoorthy *et al.* (2014) [11], Jayapriya and Bhagyalakshmi, (2016) [9] and Mamatha and Thangavel, (2018) [13]. It was found that, carbohydrates, phenolic compounds, flavonoids, alkaloids and proteins were present in copious amounts and less amounts of glycosides, saponins and steroids were present in the extract.

Muthukumar and Veerappa (2018) [16] reported presence of steroids, terpenoids, esters, acids and tannins upon phytochemical analysis of methanolic extracts of *T. populnea* leaves. *Thespesia populnea* is a herbal medicinal plant known for its therapeutic value in inflammatory disorders (Muthukumar and Veerappa, 2018) [16]. The strong antibacterial and antifungal activity was due to the presence of tannins (Ramesh, 2016) [18]. Laxmi and Pranita, (2012) [12] opined that *T. populnea* extracts possessed antioxidant capacity, which is attributed to the presence of flavanoids. Phenolic compounds possess biological properties such as

anti apoptosis, antiaging, anti inflammation, anti atherosclerosis, cardiovascular protection, improvement of endothelial function, inhibition of angiogenesis and cell proliferation activities (Han *et al.*, 2007) [7] and flavanoids have been found to be anti microbial substances against wide range of microorganisms *in vitro* (Marjorie, 1996) [15].

The presence of carbohydrates, phenolic compounds, flavanoids, alkaloids and proteins in high quantities and other compounds like glycosides, saponins and steroids at a lower

stage indicates that it is having active principles at a higher level and anti nutritional factors at a lower level. Presence of all the beneficiary constituents suggests its superiority in favourable constituents. The presence of various bioactive compounds confirms the application of methanolic extract of *Thespesia populnea* leaves for various ailments. However, isolation of individual phytochemical constituents may proceed to find a novel drug.

Table 1: Phytochemical analysis of *T. populnea* methanolic leaf extract

Phytochemical constituent	Test	Inference
Carbohydrtes	Molisch test	+++
	Fehlings test	+++
Phenolic compounds	Ferric chloride test	+++
	Lead acetate test	+++
Flavanoids	Shinoda test	+++
Alkaloids	Hager's test	+++
	Wagner's test	+++
	Mayer's test	+++
Glycosides	Legal's test	++
Saponins	Foam test	++
Steroids	Leibermann Buchards test	++
Proteins	Biuret test	+++
	Ninhydrin test	+++

+++ = Copiously present, ++ = Moderately present.

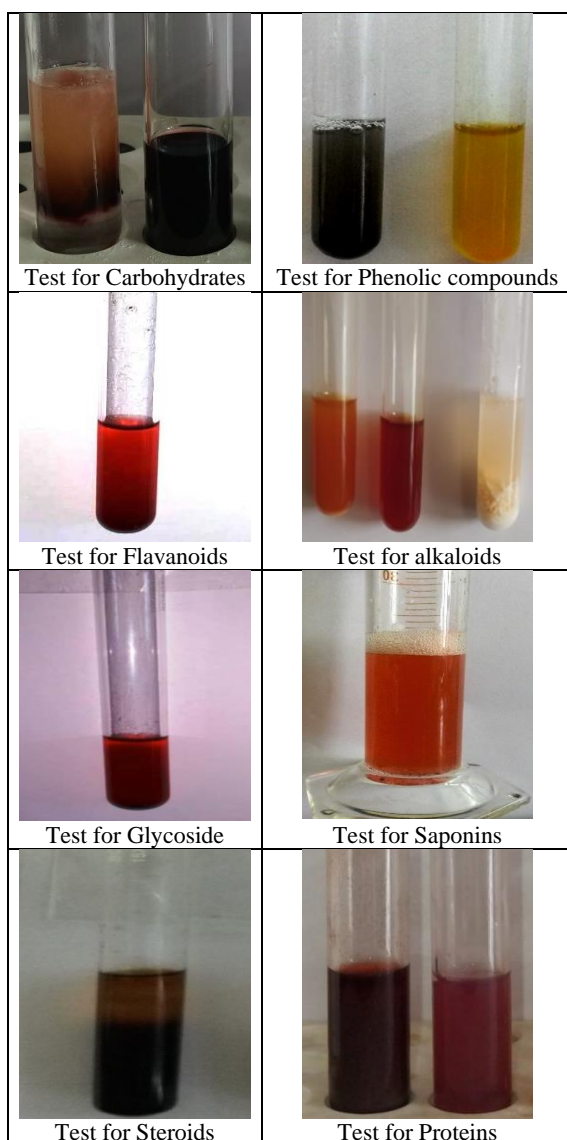


Plate 1: Phytochemical analysis of *T. populnea* methanolic extract

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