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Research Article

PHARMACOGNOSTIC AND PRELIMINARY PHYTOCHEMICAL ANALYSIS OF M*ILLINGTONIA HORTENSIS* L. AND *TECOMA STANS* L.

Karthiya.V and Vijayalakshmi A*

Department of Botany, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, Tamilnadu, India

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ABSTRACT

The present study deals with the determination of pharmacognostic features and phytochemical analysis of *Millingtonia hortensis* L. and *Tecoma stans* L. leaves belonging to the family Bignoniaceae. The phytochemical screening of ethanol, acetone, chloroform and water extracts shows the presence of carbohydrate, reducing sugar, protein, alkaloids, phenols, phytosterols and amino acid and absence of cardiac glycosides in all the extracts except chloroform extract. From this study it indicates that *Millingtonia hortensis* L and *Tecoma stans* L. have high source of secondary metabolites.

INTRODUCTION

Medicinal plants play an important role in curing various diseases in allopathic, ayurvedic and traditional medicine. For the past thousands of year medicinal plants have been used to treat health disorders and to add flavor to food.

Figure 1 *Millingtonia hortensis* L.

Millingtonia hortensis L. commonly known as cork tree are found throughout Southern Asia. It is well known for its fragrant flowers and it belongs to the family Bignoniaceae. This tree is grown as ornamental plant in gardens and avenue, The plant parts are used as antipyretic, sinusitis, cholagogue and tonic in folklore medicine (Tansuwanwong *et al.,* 2009). It is also rich in flavonoids, tannin, alkaloids and essential oil (Sharma *et al.,* 2007). Stem bark of plant is mainly used to cure lung diseases, antiasthmatic and antimicrobial (Anonymous, 2003).

Figure 2 *Tecoma stans* L.

Tecoma stans L. is an ornamental shrub belonging to the family Bignoniaceae. The flowers are traditionally used for many

Corresponding author:* **Vijayalakshmi A
Department of Botany, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, Tamilnadu, India

ailments (Kameshwaran *et al.,* 2012) and known as the national flower of the Bahamas. Plants are used to cure the diabetes and roots for urinary disorder and antifungal activity (Anburaj *et al.,* 2016). Leaves contain many bioactive compounds like alkaloids, flavonoids, steroids, tannins, phytosterols, hydrocarbons, resin, volatile oil and glycosides.

MATERIALS AND METHODS

Collection of samples

Millingtonia hortensis L. and *Tecoma stans* L. were collected from Avinashilingam Institute for Home science and higher education for women, Coimbatore, Tamil Nadu, India.

Preparation of plants extracts

The collected plants samples were thoroughly rinsed with distilled water, Afterwards the sample were dried under shade and ground. 10g of powder was soaked in 50ml of each distilled water, ethanol, acetone and chloroform for 24 hr kept in orbital shaker and then filtered through Whatman No.1 filter paper.

Pharmacognostic study

The pharmacognostic studies of *Millingtonia hortensis* L. and *Tecoma stans* L. leaves were evaluated using the following organoleptic study and fluorescence analysis. This studies were done according to the standard procedure (Jackson and Snowdown, 1968; Kokshi *et al.,* 1958; Chase and Pratt, 1949).

Phytochemical Screening

Test for Carbohydrates

Molisch's test: To a small amount of the extract few drops of Molisch's reagent was added followed by the addition of conc. H_2SO_4 along the sides of the test tube. The mixture was then allowed to stand for 2 min and then diluted with 5 ml of distilled water. Formation of red or dull violet colour at the inter phase of two layers indicates the presence of carbohydrates.

Test for Reducing Sugar

Benedict's Test: Extract (2 ml) were treated with 2 ml of Benedict's reagent and heated in a water bath for 3 minutes. Presence of green, red or yellow precipitate indicates the presence of reducing sugar.

Test for Proteins

Biuret test: To the extract (3 ml) few drops of 10% sodium chloride and 1% copper sulphate was added results in formation of violet or purple colour. On addition of alkali, it becomes dark violet.

Test for Alkaloids

Mayer's test: Sample (2 ml) was treated with few drops of Mayer's reagent. Appearance of white precipitate indicated the presence of alkaloids.

Wagner's test: Sample (2 ml) was mixed with few drops of Wagner's reagent. Appearance of reddish brown precipitate indicated the presence of alkaloids.

Hager's test: Sample (1 ml) solution and few drops of Hager's reagent were added. Appearance of yellow precipitate indicated the presence of alkaloids.

Tests for Flavonoids

Shinoda test: Sample extract was treated with 5 ml of 95% ethanol, few drops of concentrated hydrochloric acid and 0.5 g of magnesium turnings were added. Pink colour was observed. Addition of increasing amount of sodium hydroxide to the residue shows yellow coloration, it decolorize after addition of acid indicates the presence of flavones.

Flavanones: Sample extract (1 ml) was taken and 10 % of sodium hydroxide was added. Yellow to orange colour formation indicates the presence of flavanones.

Alkaline test: Sample extract (1 ml) was treated with few drops of sodium hydroxide. Yellow colour is formed which turns to be colourless after adding a few drops of dilute HCl.

Test for Glycosides

Bromine water test: Sample (1 ml) was treated with 3 drops of bromine water and the formation of yellow precipitate indicates the presence of glycosides.

Legal's test: Sample extract was mixed with few drops of pyridine and 2 drops of 2 % sodium nitroprusside. To the reaction mixture 0.5 ml of 20 % sodium hydroxide was added. Appearance of pink to red color indicated the presence of glycosides.

Test for Cardiac Glycosides

Keller-killani test: Five ml of extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was under layered with 1 ml of conc. Sulphuric acid. A brown ring at the interface indicated the presence of cardiac glycoside.

Test for Anthroquinone

Borntragor's test: Extract (0.5 ml) was added with 5-10 ml of dilute hydrochloric acid and boiled on water bath for 10 minutes. Solution was filtered and filtrate was extracted with benzene and mixed with ammonia solution. Red color was obtained in ammonia layer that indicated the presence of anthroquinone.

Test for Terpenoids

Salkowki's test: Few drop of extracts were treated with chloroform (0.5 ml) and 1ml of Conc. H_2SO_4. Formation of reddish brown precipitate shows the presence of terpenoids.

Test for Saponin

Foam test: To 1 ml of the extract 5 ml of distilled water was added and shaken vigorously. Formation of foam indicated presence of saponins.

Test for phenols

Extract (2 ml) was treated with 3ml of 10 % lead acetate. Formation of precipitate indicates the presence of phenols.

Test for Tannins

To 1ml of extract solution, 4 ml of water and 1-2 drops of 10 % ferric chloride solution was added. Blue colour indicates gallic tannins and green black indicates catecholic tannins.

Test for Catechin

Match stick was dipped in plant extract, dried and then moistened with concentrated HCl. Warm near flame, a red or pink wood is produced which shows the presence of catechin.

Test for Phlobatanins

0.5 g extract was dissolved in distilled water and filtered. The filtrate was boiled with 2M HCl solution. Formation of red precipitate showed the presence of phlobatanins.

Test for Quinones

One ml of test solution is added with alcoholic KOH solution. Quinones were indicated by colour ranging from red to blue.

Test for Sterols

Extract (2 ml) was treated with 2 ml of trichloroacetic acid. On heating the colour changes from red to violet. This indicates the presence of sterols.

Test for Phytosterols

0.2g of the extract was mixed with 2 ml of chloroform and concentrated 6M sulphuric acid (3ml) was carefully added forming a layer. A reddish brown coloration of the interface indicate the presence of phytosterols.

Test for Coumarins

Extract (2ml) was treated with 3 ml of 10% NaOH (filter paper dip in 10% NaOH solution) and 2 minutes for water bath. Coumarins is indicated by green fluorescence and examined under UV light.

Test for Amino acid

Extract (3 ml) was mixed with 3 drops of 5% ninhydrin solution and heated in boiling water bath for 10 minutes. Purple colour indicated the presence of amino acid.

Test for Diterpenes

Copper acetate test: 3 ml extract was mixed with 10 drops of copper acetate solution, Appearance of emerald green indicated the presence of diterpenes.

Test for Resin

Extract (3 ml) was mixed with 5 ml of distilled water and formation of turbidity indicated the presence of resin.

Test for steroids

2 ml of extract mixed with 2 ml of acetic anhydride solution and followed by the addition of conc. H_2SO_4 along the sides of the test tube. Violet or blue-green colour indicates the presence of steroids.

RESULTS AND DISCUSSION

Organoleptic study

The results of organoleptic study revealed the characters such as colour, odour and taste of the powder of *Millingtonia hortensis* L. and *Tecoma stans* L. were presented in table 1. The colour of dried leaf powder of *Millingtonia hortensis* L. and *Tecoma stans* L. was dark green and light green respectively. The odour and taste of the leaf powder of *Millingtonia hortensis* L. and *Tecoma stans* L. showed the characteristic odour and bitter taste.

Table 1 Organoleptic analysis of *Millingtonia hortensis* L. and *Tecoma stans* L. powder**.**

Leaf	Colour	Odour	Taste
Millingtonia hortensis	Dark green	Characteristic	Bitter
Tecoma stans	Light green	Characteristic	Bitter

Fluorescence analysis

The fluorescence property of *Millingtonia hortensis* L. and *Tecoma stans* L. powders treated with chemical reagents under day light and UV light are summarized in Table 2.

Table 2 Fluorescence analysis of *Millingtonia hortensis* L. and *Tecoma stans* L. powder sample

S.No	Treatment of powder	Colour under day light		Colour under UV light	
		M.hortensis	***T.stans***	***M.hortensis***	***T.stans***
1	Powder as such	Seaweed	Moss	Seaweed	Pickle
2	Ammonia	Juniper	Juniper	Moss	Emerald
3	Iodine	Seaweed	Chartreuse	Seaweed	Seaweed
4	$FeCl_3$	Pear	Pear	Pine	Basil
5	H_2SO_4	Chartreuse	Amber	Seafoam	Mint
6	Ethanol	Shamrock	Pickle	Fusica	Strawberry
7	Benzene	Lime	Chartreuse	Strawberry	Punch
8	Acetic acid	Lime	Juniper	Punch	Strawberry
9	Chloroform	Moss	Lime	Strawberry	Olive
10	Petroleum ether	Seaweed	Crocodile	Pickle	Olive
11	HCl	Chartreuse	Shamrock	Pickle	Crocodile
12	HNO_3	Ginger	Apricot	Marmalade	Squash
13	Acetone	Lime	Crocodile	Punch	Rouge
14	NaOH	Green	Chartreuse	Pear	Fern

Preliminary phytochemical screening

In the present study, the phytochemical screening was performed with ethanol, acetone, chloroform and water extract in leaves of *Millingtonia hortensis* L. and *Tecoma stans* L. The results indicated both the presence and absence of different types of active constituents such as carbohydrate, reducing sugar, protein, alkaloids, phenols, flavonoids, amino acid, phytosterols, terpenoids etc., The carbohydrate, reducing sugar, protein, alkaloids, phenols, phytosterols, amino acid were present in all the extracts used for two plants. Flavonoids absent in chloroform extract and coumarins & steroids absent in water extract. Among the four extracts used anthroquinones, terpenoids, catechin, phlobatanins, quinones and sterols were present only in chloroform extracts. Ethanol and acetone extract of *Millingtonia hortensis* L. and *Tecoma stans* L. exhibited presence of tannin, diterpenes and resin. In the present investigation *Millingtonia hortensis* L. and *Tecoma stans* L. showed presence of glycosides in acetone and water extract and saponin in chloroform and water extracts (Table 3).

Similar result were obtained by (Mahesh kumar *et al.,* 2014) in presence of total phenols, flavonoids, terpenoids, glycosides and protein in chloroform and water extracts of *Millingtonia hortensis* L. followed by (Chumbhalel *et al.,* 2016) preliminary phytochemical analysis of *Millingtonia hortensis* L. stem in methanol and aqueous extract have shown presence of carbohydrate, glycosides, flavonoids and tannins. Similar finding reported by (Joselin *et al.*, 2013) in different extracts (aqueous, petroleum ether, chloroform, ethanol and acetone) of *Millingtonia hortensis* L. and *Tecoma stans* L. flowers revealed

the presence of flavonoids, saponin, glycosides, terpenoids, quinines and carbohydrate.

Table 3 Preliminary phytochemical analysis of different plant extract of *Millingtonia hortensis* L. and *Tecoma stans* L.

S.No	Name of the Phytochemicals	*Millingtonia hortensis* L.				*Tecoma stans* L.			
		E	A	C	W	E	A	C	W
1	Carbohydrates	+	+	+	+	+	+	+	+
2	Reducing Sugar	+	+	+	+	+	+	+	+
3	Protein	+	+	+	+	+	+	+	+
4	Alkaloids	+	+	-	-	+	+	-	-
		-	+	+	-	-	+	+	+
		+	-	-	+	+	-	-	+
		-	-	-	+	-	-	-	+
5	Flavonoids	+	+	-	+	+	+	-	+
		-	-	-	+	-	-	-	+
6	Glycosides	-	-	-	-	-	-	-	-
		-	+	-	+	-	+	-	+
7	Cardiac glycosides	-	-	+	-	-	-	+	-
8	Anthraquinone	-	-	-	+	-	-	-	+
9	Terpenoids	-	-	-	+	-	-	-	+
10	Saponins	-	-	+	+	-	-	+	+
11	Phenols	+	+	+	+	+	+	+	+
12	Tannin	+	+	-	-	+	+	-	-
13	Catechin	-	-	-	+	-	-	-	+
14	Phlobatanins	-	-	-	+	-	-	-	+
15	Quinones	-	-	-	+	-	-	-	+
16	Sterols	-	-	-	+	-	-	-	+
17	Phytosterols	+	+	+	+	+	+	+	+
18	Coumarins	+	+	+	-	+	+	+	-
19	Amino acid	+	+	+	+	+	+	+	+
20	Diterpenes	+	+	-	-	+	+	-	-
21	Resin	+	+	-	-	+	+	-	-
22	Steroids	+	+	+	-	+	+	+	-

E - Ethanol A - Acetone + present
C - Chloroform W - Water - absent

The preliminary phytochemical screening of aqueous, ethanol, methanol, chloroform and petroleum ether extracts of *Millingtonia hortensis* L. leaves were carried out by (Janaki *et al.*, 2017) from their study it is confirmed that all the extracts showed the presence of flavonoids, alkaloids, cyanins, phenols, and coumarins which are similar to the (Sunitha, 2018) finding in methanol extracts of *Tecoma stans* L. which exhibited presence of alkaloids, glycosides, carbohydrates, steroids, tannin, phenols and anthocyanins, and absence of phytosterols, saponin, flavonoids, protein and betacyanin. The ethanol extract of *Tecoma stans* L. flower contain carbohydrate, amino acid, proteins, steroids, glycosides, saponin, alkaloids, flavonoids and phenolic compound & tannins (Sowjanya and Srinivasa, 2017). Similar results were obserbed by (Rohit Kumar, 2017) where methanol and aqueous extract of *Tecoma stans* L. contain saponin, alkaloids, flavonoids and phenols.

CONCLUSION

The result revealed the presence of more secondary metabolites in *Millingtonia hortensis* L. and *Tecoma stans* L. leaves. In future this result will help us to use this plants to produce new medicine with low cost and less side effects.

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