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Exploration of preliminary phytochemical studies of leaves of *Murraya paniculata* (L.)

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Abstract

Murraya paniculata Linn. (*Murraya exotica* L.) belongs to the family Rutaceae, and is commonly known as orange jasmine and Honeybush. It is distributed over the greater part of India and the Andaman Islands to an altitude of 1500 m. This study measure different phytochemical constituent in the *M. paniculata* leaves extract. The extract revealed the presence of alkaloids, flovonoids, phenolic compounds, carbohydrate, proteins & amino acids and; while fixed oil, saponins and mucilage were absent. The present study provides a scientific rationale for the traditional use of *M. paniculata* leaves and phytochemical exploration could be useful in future experimental studies.

Key-Words: *Murraya paniculata, Murraya exotica,* Phytochemical analysis, Hydro-alcoholic extract

Introduction

Murraya paniculata Linn. (Synonyms: *Chalcas paniculata* L., *Chalcas exotica* L. and *Murraya exotica* L.) belongs to the family Rutaceae, and is commonly known as orange jasmine. It is distributed over the greater part of India and the Andaman Islands to an altitude of 1500 m. Native to tropical Asia from India and Srilanka to Myanmar (Burma), southern China and Taiwan, Thailand, and east words throughout the Malesian region to northeastern Australia and Caledonia. It is a small tree with a spreading crown and short, often crooked, trunk; rather corky, fragrant. Leaves alternate, imperipinnate, 10-17 cm long; leaflets usually 3-5, mostely 3-7 cm long, ovate or elliptic-lanceolate or rhomboid, glossy and darker above, gland-doted, base cuneate or rounded. The leaves are stimulant and astringent; they are reportedly used in the form of an infusion to treat diarrhoea and dysentery in the Philipines. The powder leaves are applies to cuts to promote healing; there decoction is taken internally to treat dropsy. Among the Baigas of north eastern Madhya Pradesh, the crushed leaves are made in to a paste and mixed with molasses to make tablets that are taken orally to treat joint pain; the leaves, cooked with mustard or sesame oil along with dried ginger, are applied externally to relieve inflamed joints.

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The warm leaf paste is applied externally to promote the healing of broken bones among the Paudi Bhuinya in northern Orissa. In the Gandhamardn hills of Orissa, the leaves and twigs are boiled to make a bath that is used to relieve stomach-ache in children and rheumatic pains in adults. The leaves and root bark are sometime used to treat rheumatism, coughs and hysteria. Coumarins, murralongin, isomurralonginol isovalerate, murrangatin, minumicrolin (murpanidin), coumurrayin, toddalenone, aurapten, toddasin gardenin A, gardenin C, gardenin E and umhengerin was isolated from the leaves[1,2]. It is reported to have anti-diabetic and antioxidant[3], anti-nociceptive and anti-inflammatory[4], anti-diarrhoeal[5], oxytocic[6] and anti-fertility[7] properties.

Material and Methods

Collection of Plant

The whole plant of Murraya paniculata (Family - Ruteceae) was collected from Botanical Garden of National Botanical Research Institute, Lucknow, India.

Preparation of Hydro-alcoholic Extract

The freshly collected plant materials (4 kg) of *Murraya paniculata* were washed with distilled water and air-dried at 30 ± 20C. Then dried it in tray drier under the control conditions and powdered. The powdered plant materials (1kg) was macerated with petroleum ether to remove fatty substances, the marc was further exhaustively extracted with of 50% ethanol for 3 days (3 X 3L) and centrifugation at 10,000 rev/min. The extract was separated by filtration and concentrated on rotavapour (Buchi, USA) and then dried in lyophilizer

(Labconco, USA) under reduced pressure obtain 95.0 g of solid residue.

Preliminary Phytochemical Screening

50% ethanolic extract of *Murraya paniculata* were subjected to qualitative tests for the identification of various active constituents viz. carbohydrate, glycoside, alkaloid, amino acids, flavanoids, fixed oil, tannins, gum and mucilage, phytosterols etc. according to [8]Kokate CK 1990 and [9]Khandelwal KR 2000.

Test for carbohydrates and glycosides

A small quantity of the extract was dissolved separately in 4 ml of distilled water and filtered. The filtrate was subjected to the following testes to detect the presence of Carbohydrate and glycosides.

Molisch's test

The filtrate was treated with 2-3 drops of 1% alcoholic α-napthol solution and 2 ml of concentrated H2SO4 was added along the sides of the test tube. Appearance of brown ring at the junction of two liquids shows the presence of carbohydrates.

Fehling's test

The filtrate was treated with 1 ml of Fehling's solution A and B and heated on the water bath. A reddish precipitate was obtained shows the presence of carbohydrate.

Test for fixed oils and fates

Spot test

Small quantity of extract was pressed between two filter papers. Appearance of oil stain on the paper indicates the presence of fixed oil.

Saponification test

Few drops of 0.5% alcoholic potassium hydroxide were added to a small quantity of various extracts along with a drop of phenolphthalein. The mixture was heated on the water bath for 1-2 hours. Formation of soap pr partial neutralization of alkali indicates the presence of fixed oils and fats.

Test for proteins and free amino acid

Small quantity of the extract was dissolved in few ml of distilled water and treated with following reagents.

Millon's test

Appearance of red color shows the presence of proteins and free amino acids.

Ninhydrin reagent

Appearance of purple color shows the presence of proteins and free amino acids.

Biuret test

Equal volumes of 5% sodium hydroxide solution and 1% copper sulphate solution were added, appearance of pink or purple color shows the presence of proteins and free amino acids.

Test for saponins

Foam test

The extract was diluted with 20 ml of distilled water and it was agitated in a graduated cylinder for 15 minutes. The formation of 1 cm layer of foam shows the presence of saponins.

Test for phenolic compounds

Small quantity of the extract was taken in distilled water and test for the presence of phenolic compounds and tannins was carried out with the following reagents.

- Dilute ferric chloride solution (5% w/v) - Violet color.
- 1% solution of gelatin containing 10% sodium chloride-White precipitate.
- 10% lead aceatte solution-White precipitate.

Test for phytosterols

Small quantity of the extract was dissolved in 5 ml of chloroform separately. Then this chloroform solution was subjected to the following tests to detect the presence of phytosteroles.

Libermann-Burchard's test

The above preapared chloroform solution was treated with few drops of concentrated sulphuric acid followed by few drops of diluted acetic acid, 3 ml of acetic anhydride. A bluish green color appeared indicates the presence of phytosterols.

Salkowski reaction

To 1 ml of the above prepared chloroform solution, few drops of concentrated sulphuric acid was added. Brown color produced shows the presence of phytosterols.

Test for Alkaloids

Small quantity of the extract was treated with few drops of diluted hydrochloric acid and filtered. The filtrate was used for the following tests.

- Mayer's reagent – cream precipitate
- Dragendroff's reagent – Orange brown precipitate
- Hager's test – yellow precipitate
- Wagner's test – Reddish brown precipitate

Test for flavonoids

With aqueous NaOH solution

Small quantity of the extract was dissolved in aqueous sodium hydroxide and appearance of yellow colour indicates the presence of flavonoids.

With conc. sulphuric acid

To a small portion of extract, concentrated sulphuric acid was added. Yellow orange color was obtained shows the presence of flavonoids.

Shinoda's test

Small quantity of extract was dissolved in alcohol; to those pieces of magnesium followed by concentrated hydrochloric acid was added drop by wise and heated.

Appearance of magenta color shows the presence of flavonoids.

Results and Discussion

The extractive value was calculated and was found to be 9.5 % w/w. The extract was further examined for its physical characterization like color, odor and consistency. The color of the extract was dark green, with a semi-solid consistency. Extract had characteristic odor, showed the presence of desired phytochemicals. Phytochemical screening of *M. paniculata* extract revealed the presence of alkaloids, flovonoids, phenolic compounds, carbohydrate, proteins & amino acids and; while fixed oil, saponins and mucilage were absent (Table 1). Phytochemicals are bioactive, non-nutrient, naturally occurring plant compounds[10]. Purified alkaloids, as well as their synthetic derivatives, are usually employed as medicinal agents for effects such as analgesic, antimalarial, antiseptic and antibacterial[11] and Phenolic constituents may be responsible for immunomodulatory activity[12]. Saponins are produced by plants to stop bacterial and fungal attacks, which makes them natural antibiotics[13]. Therefore, the detection of saponins in the extracts could be a contributing factor for their antimicrobial properties[14]. The presence of tannins in the extracts might be responsible for hastening the healing of wounds and inflamed mucous membranes[15]. In general, the presence of these phytochemicals probably accounted for the much-touted medicinal efficacy of the extracts.

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Table 1: Preliminary phytochemical screening of the 50 % ethanolic extract of *M. paniculata*

S. No.	Constituents	Tests	50% Ethanolic extract
1.	Carbohydrate	Molish's test	+
		Fehling's test	+
2.	Fixed oil & fats	Spot test	−
		Saponification test	−
4.	Proteins & amino acids	Million's test	+
		Ninhydrin test	+
		Biuret test	+
5.	Saponins	Foam test	−
6.	Phenolic compunds	FeCl3 test	+
		Gelatin test	−
		Lead acetate test	+
7.	Phytosterol	Salkowiski test	−
		Libermann burchard test	+
8.	Alkaloids	Dragendroff's test	+
		Mayer's test	+
		Wagner's test	+
		Hager's test	−
9.	Gum & mucilage	Swelling test	−
10.	Flavonoids	Aqueous NaOH test	+
		Con. H_2SO_4 test	+
		Shinoda's test	+

Where, + = Presence, − =Absence