

PHYTOCHEMICAL SCREENING OF *NERIUM OLEANDER* LEAVES AND *MOMORDICA CHARANTIA* LEAVES

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ABSTRACT

The developing countries mostly rely on traditional medicines. The traditional medicine involves the use of different plant extracts or the bioactive constituents. This type of study provides the health application at affordable cost. This study such as ethnomedicine keenly represents one of the best avenues in searching new economic plants for medicine. In keeping this view in mind the present investigation is carried out in *Nerium oleander* and *Momordica charantia* leaves. Qualitative phytochemical analysis of these two plants confirms the presence of various phytochemicals like carbohydrates, cholesterol, protein, amino acid, alkaloid, flavonoids, tannins, saponins, cardiac glycosides, terpenoids, and phlobatinins in their aqueous leaf extracts leaves followed by ethanol, ethyl acetate, diethyl ether and chloroform. The results suggest that the phytochemical properties of the leaves for curing various ailments and possess potential antioxidant properties.

KEYWORDS: *Momordica charantia*, *Nerium oleander*, Phytochemical, Medicinal plant, Solvent extraction, Secondary Metabolites.

INTRODUCTION

World plant biodiversity is the largest source of herbal medicine and still about 60 -80% world population rely on plant based medicines which are being used since the ages as traditional health care systems. It is now clear that, the medicinal values of these plants lies in the bioactive phytochemical constituents that produce definite physiological effects on human body. Though the traditional Indian system of medicine has a long history of use, they lack adequate scientific documentation, particularly in light modern scientific knowledge¹. These natural compounds formed the base of modern drugs as we use today^{2,3,4}. "Phyto" is the greek word for plant. There are many families of phytochemicals and they help the human body in a variety of ways. Phytochemicals may protect human from various diseases. Phytochemicals are non-nutritive plant chemicals that have protective or disease preventive properties. Phytochemicals are basically divided into two groups that is primary and secondary metabolites; according to their functions in plant metabolism. Primary metabolites comprise common sugars, amino acids, proteins and chlorophyll while secondary metabolites consist of alkaloids, flavonoids, tannins and so on^{5,6}.

Bitter melon, *Momordica charantia* grows in tropical areas, including parts of Amazon, East Africa, Asia and the Carribean. It is a genus of annual or perennial climbers found throughout India and is also cultivated up to an altitude of 1500m³. The Latin name *Momordica* means 'to bite'. Bitter melon contains an array of novel and biologically active phytochemicals including cardiac glycosides, proteins and steroids show potential activity against antihelminthic, antileukemic, antibiotic, antiinflammatory, antitumor and various other properties^{7 and 8}.

Nerium oleander (Apocyanaceae family) is a beautiful free flowering shrub bearing different colors of flowers especially suited to sunny and dry localities⁹. Laboratory studies of oleander suggest possible anti cancer effects, although reliable research in humans has not yet been performed¹⁰. The

flowers, leaves, leaf juice, latex, bark and root have been used against corns, warts, cancerous ulcers, carcinoma, ulcerating or hard tumors.

MATERIALS AND METHODS

Plant Materials

The fully mature leaves of *Nerium oleander* and *Momordica charantia* were collected from Kovaipudur and Telugupalayam village in Coimbatore district of Tamil Nadu, India during July- August 2010. The leaves were washed thoroughly and shade dried.

Extraction of Plant Material

Aqueous Extraction

10g of air dried powder was added to distilled water and boiled on slow heat for 2hours. It was then filtered through 8 layers of muslin cloth and centrifuged at 5000g for 10 minutes. The supernatant was collected. This procedure was repeated twice. After 6 hours, the supernatant were collected at an interval of every 2 hours and was pooled together, concentrated to make the final volume one-fourth of the original volume. It was then autoclaved at 121°C and at 15lbs pressure and stored at 4°C⁵.

Preparation of Other Extracts

10gm of air dried powder were taken in 100ml of ethanol, chloroform, diethyl ether and ethyl acetate. Plugged with cotton wool and then kept on a rotary shaker at 190-220 rpm for 24 hours. After 24 hours the supernatant were collected and the solvent were evaporated to make the final volume one-fourth of the original volume and stored at 4°C in air tight containers⁵.

Preliminary Phytochemical Screening

The condensed extracts were used for preliminary screening of phytochemicals such as carbohydrates (Molisch's test) cholesterol (Lieberman Man Burchard test) protein (Biuret test) amino acid (Ninhydrin test) alkaloid (Wagner and Dragendroff's test), flavonoids, tannins, saponins, cardiac glycosides (Keller Killani test) terpenoids (Salkowski test) and phlobatinins.

Screening Procedure

Test for Carbohydrates

To 2ml of extract 2drops of Molisch's reagent was added and shaken well. 2ml of con.H₂SO₄ was added on the sides of the test tube. A reddish violet ring appeared at the junction of two layers immediately indicated the presence of carbohydrates¹¹.

Test for Cholesterol

To 2ml of the extract 2ml of the chloroform was added in a dry test tube. Then 10 drop of acetic anhydride and 2 to 3 drops of con. H₂SO₄ was added. A red rose color changed to blue green color¹¹.

Test for Proteins

To 2ml of protein solution 1ml of 40% NaoH solution and 1 to 2 drops of 1% CuSO₄ solution was added. A violet color indicated the presence of peptide linkage of the molecule¹¹.

Test for Amino Acids

To 2ml of sample added 2ml of ninhydrin reagent and kept in water bath for 20 minutes. Appearance of purple color indicated the presence of aminoacids in the sample⁶.

Test for Alkaloids

To the extract added 1% Hcl and 6 drops of Mayer's reagent and Dragendroff's reagent. An organic precipitate indicated the presence of alkaloids in the sample⁶.

Test for Flavonoids

5ml of dilute ammonia solution were added to a portion of aqueous filtrate of each plant extract followed by addition of con. H₂SO₄. A yellow coloration is observed which confirms the presence of flavonoids and it disappears on standing¹².

Test for Terpenoids

5ml of each extract was added to 2ml of chloroform and 3ml of con. H₂SO₄ to form a monolayer of reddish brown coloration of the interface was showed to form positive result for the terpenoids¹².

Test for Cardiac Glycosides

5ml of each extract was treated with 2ml of glacial acetic acid containing one drop of ferric chloride solution. This was underlayed with 1ml of con. H₂SO₄. A brown ring of the interface indicated a deoxy

sugar characteristic of cardenolides. A violet ring might appear below the brown ring whereas the acetic acid layer, a greenish ring might form just gradually throughout thin layer¹².

Test for Steroids

2ml of acetic anhydride was added to 0.5g of ethanolic extract of each sample with 2ml of H₂SO₄. The color change from violet to blue or green indicated the presence of steroids².

Test for Tannins

5ml of extract was added to few drops of 1% lead acetate. A yellow precipitate indicated the presence of tannins².

Test for Saponins

The extract with 20ml of distilled water was agitated in a graduated cylinder for 15 minutes. The formation of 1cm layer of foam indicated the presence of saponins⁶.

Test for Phlobatinins

When an aqueous extract of each plant sample were boiled with 1% aqueous Hcl, red precipitate was deposited which was taken as evidence for the presence of phlobatinins².

RESULTS

The present study carried out on the plant samples revealed the presence of medicinally active metabolites. The phytochemical characters of both the plants investigated are summarized in the Table 1 given below. The aqueous extract of both the plants were found to contain carbohydrates, proteins, amino acids, sterols, alkaloids, flavonoids, phlobatinins and terpenoids. Cardiac glycosides and saponins were present in *Momordica charantia* and was absent in *Nerium oleander*. Cholesterol was found to be absent in both the aqueous extracts.

Ethanolic extracts of the plant was found to contain carbohydrates, proteins, amino acids, alkaloids and cardiac glycosides. Cholesterol sterol and phlobatinins was found to be present in *Momordica charantia* and not in *Nerium oleander* while tannins and saponins were present in *Nerium oleander* and not in *Momordica charantia*. Flavonoids and terpenoids were absent in both the plant leaves.

Diethyl ether extract of both the plants showed presence of carbohydrates, proteins, amino acids, sterols and alkaloids. Cardiac glycosides, saponins and cholesterol were present in *Momordica charantia* and was absent in *Nerium oleander*. Flavonoids, phlobatinins, terpenoids and tannins were absent in both the plants.

Ethyl acetate extract of both the plants showed presence of proteins, aminoacids, sterols, cardiac glycosides and tannins. Cholesterol was present in *Momordica charantia* and was absent in *Nerium oleander*. Carbohydrates were present in *Nerium oleander* and not in *Momordica charantia*. Alkaloids, flavonoids, saponins, terpenoids and phlobatinins were absent in both.

Chloroform extracts of both the plants showed presence of carbohydrates, cholesterol, proteins, amino acids, sterols, cardiac glycosides and tannins. Alkaloids, flavonoids, saponins, terpenoids and phlobatinins were absent in both the extracts.

DISCUSSION

The phytochemical screening, and qualitative estimation of the plants studied showed that the leaves were rich in carbohydrates, proteins, amino acids and sterols in all the extracts. Some extracts showed presence of alkaloids and flavonoids too. Steroids were found to be present in almost all the extracts of the plants. It should be noted that steroidal compounds are of importance and of interest in pharmacy due to their relationship with such sex hormones. This may be the reason the leaves of *Momordica charantia* are used as vegetable for expectant mothers or breast feeding mothers to ensure their hormonal balance, since steroidal structure could serve as potent starting material in the synthesis of hormones.

The presence of cardiac glycosides has also been reported, by other researchers in *Momordica charantia* and this plant is widely used in Indian medicinal system. The plant studied here can be seen as a potential source of useful drugs. Further studies are going on, in these plants to identify furthermore uses in the field of medicine. The antioxidant activities of these plants for the treatment of the diseases as claimed by traditional healers are also being investigated.

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Table 1: Phytochemical screening of *Momordica charantia* and *Nerium oleander* leaves

	Name of the compound	Plant extract	Aqueous extract	Ethanol extract	Diethyl ether extract	Ethyl acetate extract	Chloroform extract
Momordica charantia	Carbohydrates	+	+	+	+	-	+
	Cholesterol	+	-	+	+	+	+
	Protein	+	+	+	+	+	+
	Aminoacid	+	+	+	+	+	+
	Sterols	+	+	+	+	+	+
	Alkaloids	+	+	+	+	-	-
	Flavonoids	+	+	-	-	-	-
	Cardiac glycosides	+	+	+	+	+	+
	Saponins	+	+	-	+	-	-
	Tannins	+	-	-	-	+	+
	Terpenoids	+	+	-	-	-	-
	Phlobatinins	+	+	+	-	-	-
	Nerium oleander	Carbohydrates	+	+	+	+	-
Cholesterol		+	-	-	-	+	+
Protein		+	+	+	+	+	+
Aminoacid		+	+	+	+	+	+
Sterols		+	+	-	+	+	+
Alkaloids		+	+	+	+	-	-
Flavonoids		+	+	-	-	-	-
Cardiac glycosides		+	-	+	-	+	+
Saponins		+	-	+	-	-	-
Tannins		+	+	+	-	+	+
Terpenoids		+	+	-	-	-	-
Phlobatinins		+	+	-	-	-	-

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