



Potential *in-vitro* phyto-chemicals investigation of leaves of *Ravenala madagascariensis* sonnerat

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ABSTRACT

Nature is the abundant source of medicines. All plants contents lots of chemicals which are call phytochemicals. These phytochemicals exhibit several biological activities. Every day we take lots of phytochemicals with foods. These phytochemicals regulate our body systems. In present investigation, the leaves methanolic extract of *R. madagascariensis* was subjected to the Preliminary phytochemicals screening. This investigation revealed that the presence of various potential phytochemicals such as tannins, saponins, steroids, terpenoids as well as triterpenoids. The presence of these phytoconstituent exhibit potential biological activities like as analgesic, cardioprotective, Lipid lowering activity, antidiabetics, hepatoprotective, antidiarrhoeal, anticancer, antiasthmatic activities, antioxidant, antimicrobial and so on. These investigation only preliminary studies to isolate potential compounds which may be use as a lead compound for several biological activities. Further depth investigation required to find out lead compounds.

Key Words: *Ravenala madagascariensis*; phytochemicals, tannins, saponins, steroids, terpenoids as well as triterpenoids



INTRODUCTION

Greek word "Phyto" indicate plant and "phytochemicals" related with plant pigments. So, routes, barks, fruits and vegetables contain bright and deep colors like as yellow, orange, red, green, blue and purple-generally those indicate several phytochemicals are available in the plant. These phytochemicals acts as nutrients and plays vital role in our daily life. We may use fruits and vegetables, whole grains, soy and nuts by eating 5-9 servings. In resent investigation more than 900 different phytochemicals have been identified in plant foods and more will be discovered. These phytochemicals are an emerging area of nutrition and health (Aiyelaagbe and Osamudiamen, 2009) and (Egwaikhide *et al.*, 2007). All phytochemicals are natural bioactive compounds found in plant foods those works with nutrients and dietary fiber to protect against disease. Recent research suggest, phytochemicals working together with nutrients found in fruits, vegetables and nuts, may help slow the aging process and reduce the risk of many

diseases including cataracts, osteoporosis, and urinary tract infections, cancer, heart disease, stroke and high blood pressure. Maximum plants enable exhibit antioxidant, analgesic, cardioprotective, Lipid lowering activity, hepatoprotective, antidiabetics, antidiarrhoeal, antidiabetics, antidiarrhoeal, anticancer, antiasthmatic activities and so on by overlapping mechanisms of action in the body (Md. Reyad-ul-ferdous *et al.*, 2014). *Ravenala madagascariensis* also called Travelers-Tree. It is Native of North America, cultivated for in many tropical and subtropical regions. It is a small spreading tree with bark does not droop; showy; typically multi-trunked; thorns belonging to the family Strelitziaceae .Tree 30-60 ft (9-18 m) tall. Simple leaf type, entire leaf margin, oblong Leaf shape, pinnate leaf venation evergreen, broad leaf evergreen leaf type, more than 36 inches length, green leaf color. Flowers are white/cream/gray. Fruit s are less than .5 inch, .5 to 1 inch length, dry or hard Fruit covering, brown In color, does not attract wildlife; not showy; fruit/leaves not a litter

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problem ("Botanical Journeys Plant Guides" 2008-2010).

MATERIALS AND METHODS

Plant material: The leaves of *R. madagascariensis* were collected from Mirpur Botanical Garden, Dhaka, Bangladesh, in January 2013. Plant identification number was collected from Bangladesh National Herbarium, Dhaka, Bangladesh (Accession no. 38302). The leaves were picked and washed with water to remove all unwanted plant materials as well as sand then air dried under light exposure (27°C-30°C for 7 days), pulverized in a mill and stored in an airtight container for further use. The air dried and powdered leaves (500 gm) of *R. madagascariensis* was macerated in 2.5 L of methanol for 7 days and then filtered through a cotton plug followed by Whatman filter paper number 1. The extracts were concentrated with a rotary evaporator at low temperature (40-45 °C) and reduced pressure. The concentrated methanolic extract (ME) was partitioned by modified Kupchan method (Van Wagenen *et al.*, 1993) and the resultant partitionates i.e., pet-ether (PESF), carbon tetrachloride (CTCSF), chloroform (CSF), and aqueous (AQSF) soluble fractions were used for the experimental processes.

Preparation of extract: The powdered plant material (200 g) was extracted thrice in distilled water (5.5 L; 27°C-30°C) on shaker (Stuart Scientific Orbital Shaker, UK) for 48 hours. The extract was filtered using a Buchner funnel and Whatman No.1 filter paper. The aqueous extract filtrate obtained was quickly frozen at -40°C and dried for 48 h using a freeze dryer (Savant Refrigerated vapor Trap, RV T41404, USA) to give a yield of 30 g of dry extract. The resulting extracts were reconstituted with distilled water to give desired concentrations used in this study.

QUALITATIVE SCREENING PROCEDURE

Phytochemicals screening of the plant extract: 1 gram of the methanol extract of *R. madagascariensis* was dissolved in 100 ml of methanol and was subjected to preliminary phytochemical screenings for determining presence and nature of phytoconstituents. In present investigation, we conduct several tests to determine phytochemicals include tannins, flavonoids, alkaloids, saponins, Terpenoids, Triterpenoids as well as steroids in accordance with the methods with little modifications (Harbone, 1998). In present study, found several potential phytoconstituents table-1. Phytochemicals was identified by characteristic color changes.

Anthraquinone (Borntrger's Test): 0.5 g of the extract was taken into a dry test tube and 5 mL of chloroform was added then shaken for 5 min. The extracts were filtered and the filtrate was shaken with equal volume of 10% ammonia solution. A red or pink violet color in the ammonical layer was observed, indicates presence of anthraquinone.

Tannins: Small amount of extract was mixed with distilled water and heated on H₂O bath. It was filtered and Ferric chloride was added to the filtrate. Dark green color indicates presence of tannins. Consequently, 5ml of extract and a few drops of 1% lead acetate were added. Yellow precipitate was formed, indicates the presence of tannins.

Flavonoids: 0.2 g was dissolved in diluted NaOH and HCl was added. Yellow solution that turns colorless indicates the presence of flavonoids.

Saponins: 0.2 g of plant extract was taken and 5 mL of distilled water was added and then boiled. Frothing persistence indicates presence of saponins.

Steroids (Liebermann- Burchard Reaction): 1ml plant material in 10 mL chloroform filtered. 200 milliliter of acetic anhydride was added to 2 mL filtrate with 2 mL H₂SO₄. The color changes from violet to blue or green in some samples indicating the presence of steroids. 1 ml of the extracts was dissolved in 10ml of chloroform and equal volume of concentrated H₂SO₄ was added by sides of the test tube. The upper layer turns red and H₂SO₄ layer showed yellow with green fluorescence which indicates the presence of steroids.

Phlobatanins: 0.5 g of plant extract was dissolved in distilled water and filtered. The filtrate was boiled with 2% HCl solution and red precipitate shows, the presence of phlobatanins.

Terpenoids (Salkowski Method): 0.5 g of extract added in 2 mL of chloroform filtered. Then Concentrated H₂SO₄ carefully added to form a layer. Reddish brown coloration of the interface was formed to show positive results for the presence of terpenoids.

Triterpenoids: 10 mg of the extract was dissolved in 1 ml of chloroform; 1ml of acetic anhydride was added following the addition of 2 ml of conc. H₂SO₄. Formation of reddish violet colour indicates the presence of triterpenoids.

Cardiac Glycoside: 0.5g of each was treated with 2 mL of glacial acetic acid containing a drop of FeCl₃ solution. This was underlayered with 1 mL of conc. H₂SO₄. Brown ring obtained at the interface indicated the presence of de-oxy sugar characteristics of cardenolides.

RESULTS AND DISCUSSION

The result obtained in the present investigation phytochemicals screening of the methanol extract of leaves used in the *R. madagascariensis* study

revealed that the crude extracts contained tannins, saponins, steroids, terpenoids and triterpenoids (Table-1). *R. madagascariensis* leaves have various medicinal values such as anti-inflammatory, antibacterial, antioxidant as well as thrombolytic activity due to presence of those potential phytochemicals. We can isolate pure lead compound from this plant for future drug development. Other potential phytochemicals also present in this plant, which exhibit several biological activities. This is only a preliminary study of the occurrence of certain properties of *R. madagascariensis* leaves an in-depth study will provide a good concerted base of all the phytochemicals functions mention above.

CONCLUSION

In the present study, we have found that most of the biologically active phytochemicals were present in the methanolic extracts of *R. madagascariensis* leaves. This is only a preliminary study and to make final comment, the extract should thoroughly investigated phytochemically and pharmacologically to exploit their medicinal and pharmaceutical potentialities.

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Conflict of Interest: All authors were declared no conflict of interest.

Table-1: Phytochemicals investigation of leaves of *R. madagascariensis*.

Name of the Test	Color	Results
Anthraquinone (Borntrger's Test)	pink violet or red color	-
Tannins	dark green color	+
Flavonoids	yellow solution that turns colorless	-
Saponins	Frothing persistence	+
Steroids (Libermann-Burchard Reaction)	The upper layer turns red and H ₂ SO ₄ layer showed yellow with green fluorescence	+
Phlobatanins	Red precipitate	-
Terpenoids (Salkowski Method)	Formation of reddish violet colour	+
Triterpenoids	H ₂ SO ₄ . Formation of reddish violet colour	+
Cardiac Glycoside	brown ring obtained	-

(+) = present and (-) = absence

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