Preliminary Phytochemical Analysis and Total Phenolic Content of Leaf Extract Rhodomyrtus Tomentosa

Kousalya. J[1], Chitra. P[2]
[1, 2] Dept of Biochemistry
[1, 2] Sri Ramakrishna College of Arts and Science for Women Coimbatore – 44, Tamil Nadu, India.

Abstract-*Medicinal plants are widely used in non-industrialized societies, mainly because they are readily available and cheaper than modern medicines. The annual global report value of 50,000 to 70,000 plants with suspected medicinal properties was estimated to be US$ 2.2 billion in 2012 [1] and in 2017, the potential global marked for botanical extracts and medicines was estimated at several hundred billion dollars [2]. In many countries, there is little regulation of traditional medicine, but the world health organization coordinates a network to encourage a safe and rational usage. Medicinal plants face both general threats such as climate change and habitat destruction [3]. Rhodomyrtus tomentosa is a member of the Myrtaceae plant family native to Indian Sub- continent, China and South-East Asia. The plant is mainly growing along coastal shores, wetlands and riparian zones, from sea level up to 2400 m elevation [3]. R.tomentosa extract possesses potential anti-inflammatory and antiulcer activity, and can serve as a potent antioxidant [4],[5],[6]. Therefore the aim of the study is to investigate the phytoconstituent and thin layer chromatography from leaves of R. tomentosa.*

Keywords- medicinal plants, phytoconstituents,phenol.

I. INTRODUCTION

Medicinal plants are the core of traditional medicine among the rural dwellers worldwide since the very beginning of civilization. The therapeutic use of herbs dates back to the third millennium BC during the Sumerian and the Acadian civilizations and are still utilised to treat numerous aliments in the traditional Chinese, Egyptian, Ayuevedic, unani, siddha and medicinal systems[7].The nature hosts with more than 2000 species of plants whose phytochemical properties are still unveiled[8].

Nature has been a source of medicinal agents for thousand of years and an impressive number of drugs have been isolated from natural sources. World plant biodiversity is the largest source of herbal medicines and still about 60-80% world population relies on plant based medicines which are being used since the ages as traditional health care system[9].

Rhodomyrtus tomentosa is a member of the Myrtaceae plant family native to Indian Sub- continent, China and South-East Asia. The plant is mainly growing along coastal shores, wetlands and riparian zones, from sea level up to 2400 m elevation[3]. The plants can grow up to 1-2 m tall, and sometimes 3 m in height. The leaves are oval in shape, rounded tips. The flowers have five bright pink petals that fade to pale pink as they age. It has five sepals at the base and numerous stamens in the center. The fruits are edible, oblong-shaped berry, and crowned by the persistent sepals[10]. Previous studies on this plant had show antibacterial activity of ethanolic leaves extract against *Streptococcus pyogenes*[11]. *R.tomentosa* extract possesses potential anti-inflammatory and antiulcer activity, and can serve as a potent antioxidant [4], [5], [6].Therefore the aim of the study is to investigate the phytoconstituent from leaves of *R. tomentosa* .

II. MATERIALS AND METHOD

PHASE I

1.1 Collection of samples
1.2 Preparation of plant extract

PHASE II

2.1 Qualitative phytochemical screening
2.2 Qualitative analysis- Total phenolic content

PHASE I

1.1 COLLECTION OF PLANT MATERIAL:

Fresh leaves of *Rhodomyrtus tomentosa* were cut and washed under running tap water to remove all foliar contaminants, dried at room temperature for 10-15 days, and pulverized to fine powder. The powdered leaves were

weighted separately and stored in container for further experiments.

1.2 PREPARATION OF EXTRACT:

The plant extract was carried out by soxhlet's method by using 1000ml in 200 grm of methanol. And the extract was kept in incubator for 24 hours. The extract was collected in petri plate for further use.

PHASE II

2.2 SCREENING OF PHYTOCHEMICAL ANALYSIS:

A systematic and complete study of crude drugs should include a complete investigation of both primary and secondary metabolites derived from plant metabolism. The different qualitative chemical tests are to be performed for establishing profiles of the extract for their nature of chemical composition. The methanolic extract of the sample, obtained as above was tested for the following qualitative chemical tests for the identification of various phytochemicals.

Qualitative analysis of Phytochemical screening:

The phytochemical screening of plant extracts were carried out by the following method of [12].

a).Test for tannins;

Ferric chloride test:
To 1.0 ml of extracts is treated with 1 % ferric chloride solution.

b) Test for alkaloids:

Mayers test:
To 1.0 ml of the extract is treated with mayer's reagent.

c) Test for flavanoids:

Alkaline reagent test:
To 1.0 ml of the leaf extract is treated with a few drops of sodium hydroxide solution

d) Test for saponins:

Foam test:
Shake 1.0 ml of the extract with 2.0 ml of water.

e) Test for terpenes:

To 1.0 ml of the extract is treated with chloroform and filtered. Treat the filtrate withfew drops of concentrated sulphuric acid, shake and allow to stand.

f) Test for glycosides:

To 1.0 ml of the extract is treated with dilute hydrochloric acid and ferric chloride solution. Immersed in boiling water for 5 minutes, cool the mixture with equal volume of benzene. Separate the benzene layer and treat with ammonia solution.

g) Test for reducing sugar:

To 1.0 ml of the extract is treated with 1.0ml of benedict's reagent and heat gently.

h) Test for phenol:

To 1.0 ml of the extract to 3-4 drops ferric chloride solution.

2.3 Qualitative analysis- Total phenolic content:

Folin Ciocalteu's colorimetric assay was used for the determination of total phenols in extracts.. The reaction mixture contains 0.3 ml (1 mg/ml) of extract, 1.5 ml of 10% Folin-Ciocalteu's reagent dissolved in water and 1.2 ml 7.5% NaHCO3. The samples were incubated for 30 min and the absorbance was determined at λmax = 765 nm. Blank was concomitantly prepared with ethanol instead of extract solution .The samples were prepared in triplicate and the mean value of absorbance was obtained. The total phenolic content was expressed in terms of gallic acid equivalent (mg of GaA/g of extract). Total phenol content was estimated by the method of [13].

III. RESULT AND DISCUSSION

PHASE: 1

SCREENING OF PHYTOCHEMICAL COMPONENTS

The methanolic extract of *R.tomentosa* leaves were subjected to phytochemical screening to test the presence of secondary metabolites such as tannins, alkaloid, flavonoid, saponins, terpenes, glycosides, reducing sugar and phenol.

In the present study secondary metabolites in the methanolic extracts of *R.tomentosa* leaves were qualitatively analyzed, using standard methods. The leaves extract demonstrated the occurrence of phytoconsituents such as

tannins, alkaloids, reducing sugar, flavonoids, and absence of saponins, terpenes, glycosides were observed.

The presence of flavonoid indicates the natural occurring phenolic compounds which is beneficial effects in human diet as antioxidant and neutralizing free radicals [14].

Table 1: Phytochemical analysis to methanolic extract of leaves of *Rhodomyrtus tomentosa*

Biochemical tests	Tests	Leaves extract
Tannins	Ferric chloride	+
Alkaloids	Mayer's test	+
Saponins	Foam test	–
Terpenes	Liebermann's	–
Glycosides		–
Reducing sugar	Benedicts test	+
Phenol	Ferric chloride	+
Flavanoid	Alkaline test	+

From the Phytochemical screening of *R.tomentosa* of methanolic extract of the leaf shows the presence of most of the phytochemical such as tannins, alkaloids, reducing sugar, phenol and flavonoid.

Preliminary phytochemical screening of the leaf extract of *Rhodomyrtus tomentosa* revealed the presence of various chemical compounds such as alkaloids, tannins, reducing sugar, phenol, flavonoid and carbohydrate.

QUALITATIVE ANALYSIS OF TOTAL PHENOLIC CONTENT:

Phenols are very important plant constituents because of their scavenging ability on free radicals due to their hydroxyl groups. Therefore, the phenolic content of plants may contribute directly to their antioxidant action [15]. Total phenolic posses a broad spectrum of chemical and biological activities including radical scavenging properties. Such property is especially distinct for flavonols .

A number of articles have publicized that plants pocess potent antioxidants which act as inhibitors of lipid peroxidation ans scavengers of free radicals in the form of phenolic compounds, vitamins and flavonoids [16]. Presence of such high amount of total phenols may contribute to the antioxidant activity of the prepared formulation.

Table 2; Levels of Total phenolic content

Name of the extract	Total Phenol (mg gallic acid/100 ml
Ethanolic extract of *Rhodomyrtus tomentosa*	82.98±1.37

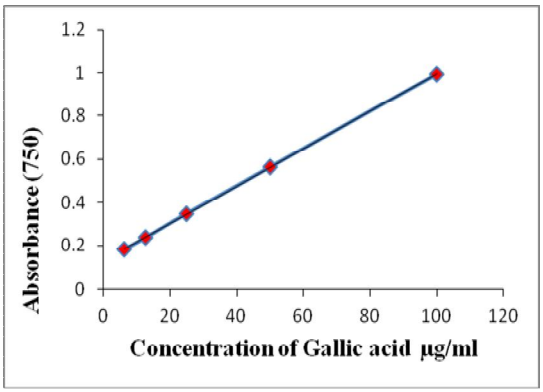

Fig 1 : Standard graph of Gallic acid

Numerous investigation of the antioxidant activity has confirmed linear correlation between the values of phenol concentration and antioxidant activity [17].

REFFERENCE

[1] "Medicinal and aromatic plants trade programme" Traffic .org. Retrived 20 February 2017.

[2] Ahn, K (2017), "The Worldwide trend of using botanical drugs and strategies for developing global drugs". **50**(3): 111-116.

[3] Csurhes, S. and Hankamer, C. (2016). *Ceylon Hill Cherry (Downy Rose Myrtle)*: *Rhodomyrtus tomentosa*. Queensland Government.

[4] Geetha, K. M., Sridhar, C. and Murugan, V. (2010). Antioxidant and Gastroprotective activities of *Rhodomyrtus tomentosa* (Ait). Hassk, *Internation Jounal of PharmTech Research*, **2**(1), 283-291.

[5] Lavanya, G., Voravuthikunchai, S. P. and Towatana, N. H. (2012). Acetone extract from *Rhodomyrtus tomentosa;* A Potent Natural Antioxidant, *Evidence-based Complementary and Alternative Medicine*, 10,1155/**2012**/535479.

[6] Jeong,D., Yang, W.S., Yang, Y., Nam, G., Kim, J.H., Yoon, D. H., Noh, H.J., Lee, S., Kim, T. W., Sung, G. and Cho, J.Y.(2013). *In vitro and in vivo* anti-inflamatoryeffect of *Rhodomyrtus tomentosa* methanolic

extract. *Journal of Ethnpharmacology*, **146**(2013), 205-213.

[7] Sala, A., Recio, M.D., Giner, R.M., Manez, S., Tournier, H., Schinella, G., Rios, J.L., Antiinflamatory and antioxidant properties of *Helichrysum italicum* .J Pharm Pharmacol 2002; 54(3): 365-371.

[8] Ramaiah, R., and Suresh, P.C., Molecular docking studies of phytochemical compounds with viral proteases, International Journal of Pharm Science Res (2013) 4(1): 475-482.

[9] Rawat RBS and Uniyal R (2003), National medicinal plant board committed for overall development of sector. Agro Bios MedPlant, 1, 12-16,(2003).

[10] Navie, S. (2013). *Ceylon Hill Cherry (Rhodomyrtus tomentosa).* Nergan: Technigro Australia.

[11] Limsuwan, S., Kayser, O. and Voravthikunchai, S. P. (2012). Antibacterial activity of *Rhodomyrtus tomentosa (*Aiton) Hassk. Leaf extract against Clinical Isolates of *Streptoccous pyogenes. Evidence-Based Complementary and Alternative Medicine,* 10. 1155/**2012/**697183.

[12] Evans.W.C and Trease. E (1997), Trease and Evans Pharmacogenosy. Harcourt Brace and Company. Asia pvt. Ltd. Singapore.

[13] Folin O, Ciocalture V (1927), On tyrosin and tryptophan determination in proteins, Journal of Biology and Chemistry, 27: 627-650.

[14] Del-Ri A, Obdululio BG, Casfillio J, Marin FG, Ortuno (1997), A Uses and properties of Citrus flavonoids. J. Agric. Food Chem; 45:4505-4515.

[15] Tosun M, Ercisil, S, Sengul M, Ozer H, Polat T (2009), Antioxidant properties and total phenolic content of eight *Saliva species* species from Turkey. *Biol.Res*. **41,** 175-181.

[16] Sharma A., Shankar. C., Tyagi, L., Singh, M., Rrao, C (2008) *Herbal medicine for Market Potential in India; An Overview*. Academic journal of plant sciences, Vol2, 26-36.

[17] Borneo R, Leon E. A., Aguirre A, Ribotta, p., Cantero, J.J. (2008), Antioxidant capacity of medicinal plants from the Province of Cordoba (Argentina) and their in vitro testing in model food system. *Food Chem.***112,** 664-670.