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Phytochemical Investigation of *Erythrina variegata* and *Ficus racemosa* leaves

M. Ramila Devi and A. Manoharan

Department of Plant Biology and Plant Biotechnology, Presidency College, Chennai

ABSTRACT

*The traditional medicine involves the use of different plant extracts or the bioactive constituents. The study such as ethno medicine keenly represents one of the best avenues in searching new economic plants for medicine. This type of study provides the health application at affordable cost. The present study carried out to find out the photochemical constituents in the *Ficus racemosa* leaves, and *Erythrina variegata* leaves, the *Ficus racemosa* and *Erythrina variegata* leaves was collected from the Presidency campus Chennai, Tamilnadu. The shadow dried bark materials were grained and extracted with benzene, ethanol, ethyl acetate, methanol and petroleum ether. Phytochemical screening was carried out according to standard procedures. Sugar, protein, alkaloids, flavonoids, sterols and glycoside were found to be present in the extracts.*

KEY WORDS: *Ficus racemosa*, and *Erythrina variegata* leaves, Phytochemical.

INTRODUCTION

Since time immemorial man has used various parts of plants in the treatment and prevention of many ailments (Chah et al., 2006). Historically all medicinal preparations were derived from plants, whether in the simple form of plant parts or in the more complex form of crude extracts, mixtures, etc. Today a substantial number of drugs are developed from plants (Fabricant, et al., 2001.) Which are active against a number of diseases? The majority of these involve the isolation of the active ingredient (chemical compound) found in a particular medicinal plant and its subsequent modification. In the developed countries 25 percent of the medical drugs are based on plants and their derivatives (Principe, 2005) and the use of medicinal plants is well known among the indigenous people in rural areas of many developing countries.

Phytochemicals may protect human from a host of diseases. Phytochemicals are non-nutritive plant chemicals that have protective or disease preventive properties (Ahmed, et al., 2010) Plant produces these chemicals to protect itself but recent research demonstrates that many phytochemicals can protect humans against diseases. There are many phytochemicals in fruits and herbs and each works differently. (Argal, and Pathak, 2006.)

Ficus racemosa Linn belonging to the family Moraceae is commonly known as 'Jagyadumur' (Bengali), 'Gular' (Hindi) and 'Udumbara' (Sanskrit) is a well-known moderate sized to large spreading tree with ovate, ovate-lanceolate leaves. The leaves are used in dysentery, diarrhoea, bilious affection and in dysmenorrhea; barks and fruits are also used in dysentery, diarrhoea and in diabetes (Chopra *et al.*, 1958; Kirtikar and Basu, 1975; Nadkarni *et al.*, 1976). The antidiarrhoeal hypoglycemic activity (Mandal *et al.*, 1997b), anti-inflammatory activity (Mandal *et al.*, 1998a), hepatoprotective activity (Mandal *et al.*, 1998b, 1999) and the treatment of bronchitis (Mandal *et al.*, 2000) are properties of the leaf extract of *F. racemosa* have been reported .

Erythrina variegata Linn. Var. belonging to the family Fabaceae is commonly known as 'Palitamadar' (Bengali), 'Pharahada, Pangara' (Hindi), 'Paribhadra, Kantakimsuka' (Sanskrit) is a medium-sized deciduous small tree with prickly stems and branches, leaves with triangular leaflets and large coral red flowers and grows all over Bangladesh. Different parts of *E. Variegata* have used in traditional medicine as nervine sedative, febrifuge, anti-asthmatic and antiepileptic. (Anwar *et al.*, 2006.) The leaves are used in fever, inflammation and joint pain. The juice of the leaves is used in earache, toothache (Ghani, 1998) constipation, (Anonymous.2002) cough (Ghosal , *et al.*, 1972.) and also known to stimulate lactation and menstruation. Leaves and Juice are being used in the traditional system of medicine for the treatment of various ailments such as liver trouble, convulsion, arthritis, etc. (Nadkarni, *et al.*, 1992; Kiritikar, *et al.*, 1991). Hence the present study has been made to investigate the phytochemical screening of the *Ficus racemosa* and *Erythrina variegata* leaves.

EXPERIMENTAL SECTION

Plant materials

Ficus racemosa and *Erythrina variegata* barks are collected from Presidency college campus Chennai, Tamil Nadu. They were washed with tap water, rinsed with distilled water and cut in to small fragments and shade dried until the fracture is uniform and smooth. Then the dried plant material was powdered. Then the final uniform powder was used for the extraction of active constituents of the plant.

Preparation of extracts

The dried powder material was extracted and dematerialized water successively in a Soxhlet apparatus. The extracts were filtered while hot and concentrated under reduced pressure. The practical and % yields of the ex- tracts were calculated. Different types of polar solvents like Ethanol Methanol [High polar solvent], Ethyl acetate, and Petroleum ether [moderate], Benzene, [low polar solvent] were used for the extractions. These extracts were used for the detection of photochemical analysis and Thin Layer Chromatography (TLC).

Pharmacognostic Evaluations**Determination of Ash**

Weigh 2-4 g of the sample accurately in previously ignited and tarred silica dish. Spread the material evenly and ignite in a muffle furnace at 600°C until it is white, indicating absence of carbon. Cool the dish in desiccators and weigh. If carbon free ash cannot be obtained in this manner, cool the dish and moisten then residue with about 2ml of water or a saturated solution of Ammonium nitrate. Dry on a water-bath, and then ignite in the muffle furnace to constant weight. Calculate the percentage of total ash of air-dried material.

Determination of Acid insoluble ash

The ash obtained as described in the determination of total ash. To the dish containing the total ash, add 45ml of 1:5 HCL in three portions of 15ml each time boil gently for 5 minutes and filter. Collect the insoluble matter on an ash less filter paper [What man no.41] and wash with distilled water until the residue is free from acid. Transfer the filter paper containing the insoluble matter to the original dish, dry and ignite to constant weight. Cool the dish in a desiccator, and then weigh. Calculate the percentage of Acid insoluble- ash of the air-dried material.

Alcohol Soluble extract

Weigh accurately 4g of the sample in a glass stoppered flask. Add 100ml of alcohol [approximately 95%] shake occasionally for 6hrs and then allow stand for 18hrs. It was then filter rapidly taking care not to lose any solvent and pipette out 25ml of the filtrate in pre-weighed 100ml beaker and evaporate to dryness on a water bath 105°C for 6hrs, cool in a desiccator for 30 minutes and weigh. Calculate the percentage of alcohol extractable matter of the sample.

Water Soluble extract

Weigh accurately 4g of the sample in a glass stoppered flask. Add 100ml of chloroform water [95ml of water+5ml of Chloroform] shake occasionally for 6hrs and then allow to stand for 18hrs. It was then filter rapidly taking care not to lose any solvent and pipette out 25ml of the filtrate in a pre-weighed 100ml beaker and evaporate to dryness on a water bath 105°C for 6hrs, cool in a desiccator for 30 minutes and weigh. Calculate the percentage of alcohol extractable matter of the sample.

TLC(Thin layer chromatography)

4g of the samples were soaked in 40ml of Ethyl alcohol and kept overnight, boiled for ten minutes and filtered. The filtrate was concentrated to 10ml and made up to the mark in a 10ml standard flask. 15µl of this solution was applied on Merck Aluminum plate 60 F254 pre-coated with silica gel of 0.2mm thickness and the plates were developed in Toluene: Ethyl acetate: Formic acid 5:1.5:0.5. After drying the plate was visualized under UV 254 and 366 nm. The plates were then dipped in Vanillin- Sulphuric acid reagent and kept in oven 105° C till the colour of the spots appeared.

Qualitative phytochemical analysis**Test for Alkaloid**

Five ml of extract was taken in acetic acid and 2ml of freshly prepared Dragendroff's reagent to be added. An orange or red precipitate shows the presence of alkaloids.

Test for triterpenoids [Noller's Test]

One ml of extract with tin and thionyl Chloride[1ml] were added. Heat in a water- bath. Purple colour shows the presence of triterpenoids.

Test for Steroids [Liebermann – Bur chard's Test]

One ml of extract 0.5ml of Chloroform, 5ml of acetic acid, heat, add 5ml of acetic anhydride and add conc. Sulphuric acid. Green Colour shows the presence of Steroids.

Test for Flavonoids [Shinadow's Test]

One ml of extract, 5-10drops of dilute HCL was added followed by a small amount Mg and the solution was boiled in a water- Bath for a few minutes. Mejanya colour shows the presence of Flavonoids.

Test for Carbohydrates [sugar]

Extract was treated with anthrone and Conc. Sulphuric acid. Heat in a water- bath. Green colour shows the presence of sugar.

Test for Quinones

One ml of extract 1ml of Conc. Sulphuric acid was added. Formation of red colour shows the presence of Quinones.

Test for Phenols

One ml of the extract, 2ml of distilled water was added followed by few drops of 10% aqueous ferric chloride. Appearance of blue or green colour indicates the presence of phenols.

Test for Saponin

One ml of extract was diluted to 5ml of water was added and the tube was shaken vigorously. Formation of honey comb, like froth indicates the presence of saponins.

Test for Tannin

One ml of extract was diluted to 5ml with distilled water in a tube and to this a few drops of lead acetate solution (1%) was added. A white precipitate indicates the presence of tannin.

RESULTS AND DISCUSSION

Herbal extracts contain different phytochemicals with biological activity that can be of valuable therapeutic index. Much of the protective effect of fruits and vegetables has been attributed by phytochemicals, which are the non-nutrient plant compounds. In the present investigation, characteristics of pharmacognosical significance of *Ficus racemosa* and *Erythrina variegata* leaves shows, total ash, acid insoluble ash, loss on drying, Alcohol soluble extractive value, water soluble extractive value, determined. The standardization values of *Ficus racemosa* leaf had relatively low on *Erythrina variegata* is given in Table-1. *Ficus religiosa* is reported to have numerous therapeutic uses in folk medicine. Leaf juice has been used for the treatment of asthma, cough, sexual disorders, diarrhoea, haematuria, ear-ache and toothache, migraine, eye troubles, gastric problems and scabies; leaf decoction has been used as an analgesic for

toothache.[Damanpreet Singh, , *et al.*, June 2009,] Fresh and dried fruit of *F.carica* is used in cancer,carcinoma, ulcers, hepatomegaly, spleenomegaly.

Table 1- The analysis of Standardization in the *Ficus racemosa*& *Erythrina variegata* leaf

S.No	Parameters	<i>Ficus racemosa</i>	<i>Erythrina variegata</i>
1.	Ash(% w/w)	11.56	9.47
2.	Acid insoluble ash (% W/W)	3.52	0.35
3.	Water –soluble extractive (% w/w)	10.89	20.6
4.	Alcohol–soluble extractive (% w/w)	1.78	4.57
5.	Loss on drying at 105°C(%w/w)	11.70	14.28

Table 2 - The analysis of phytochemicals in the different organic extracts of *Ficus racemosa* and *Erythrina variegata* leaves

Phtochemicals	Leaf inference									
	<i>Ficus racemosa</i>					<i>Erythrina variegata</i>				
	B	E	EA	M	PE	B	E	EA	M	PE
Steroid	+	+	+	+	+	+	+	+	+	±
Triterpenoid	+	+	+	+	+	+	-	+	-	+
Flavonoid	+	+	+	+	-	+	-	+	-	+
Furan	-	+	-	+	+	-	-	+	-	-
Sugar	+	+	+	+	+	+	-	-	-	+
Coumarin	+	+	+	+	-	-	+	+	+	-
Quinine	-	+	-	+	+	-	-	-	-	-
Alkaloid	+	-	+	-	+	+	+	+	+	+
Tannin	+	-	+	-	+	-	-	+	-	-
Phenol	+	+	+	+	+	+	+	+	+	+
Acid	-	-	-	-	-	-	-	-	-	-
Saponin	+	-	+	-	+	+	+	+	+	-

+ = presence; - = Absence; B = Benzene; E = Ethanol; EA = Ethyl acetate; M = Methanol; PE = Petroleum ether.

Table 3a - TLC of *Ficus racemosa* leaf

S.No	UV254 nm		UV366nm		With spray reagent	
	Colour	Rf	Colour	Rf	Colour	Rf
1.	Green	0.16	Red	0.16	Pale brown	0.16
2.	Green	0.79	Red	0.24	Pale brown	0.41
3.			Red	0.29	Grey	0.64
4.			Blue	0.34	Grey	0.56
5.			Red	0.59	Pale grey	0.60
6.			Red	0.69	Grey	0.64
7.			Red	0.81	Grey	0.68
					Grey	0.74
					Pale brown	0.79
					Grey	0.96

Latex is used in ulcers and gout. Leaves are used in cancer, tumours, dermatitis. [Anwarul Hassan Gilani, *et al.*, 2008,].Different phytochemicals have been found to possess a wide range of activities, which may help in protection against chronic diseases. For example, glycosides,

saponins, flavonoids, tannins and alkaloids have hypoglycemic activities, anti-inflammatory activities (Augusti *et al.*, 2008) (Rupasinghe, *et al.*, 2003) reported that saponins possess hypocholesterolemic and antidiabetic properties. The terpenoids have also been shown to decrease blood sugar level in animal studies (Luo, *et al.*, 1999). Steroids, triterpenoids and saponins showed the analgesic properties and central nervous system activities. (Sayyah, *et al.*, 2004; Malairajan, *et al.*, 2006) are shown in Table -2. TLC *Ficus racemosa* and *Erythrina variegata* leaves are shown in plate I and Table -3a and 3b.

Table 3b - TLC of *Erythrina variegata* leaf

S.No	UV254nm		UV366nm		With spray reagent	
	Colour	Rf	Colour	Rf	Colour	Rf
1.	Green	0.27	Blue	0.01	Grey	0.15
2.	Green	0.78	Red	0.15	Grey	0.37
3.			Red	0.19	Grey	0.51
4.			Red	0.22	Brown	0.60
5.			Red	0.29	Grey	0.66
6.			Red	0.57	Grey	0.72
7.			Red	0.66	Brown	0.79
8.			Red	0.72	Brown	0.82
9.			Red	0.79	Brown	0.91
10.			Red	0.90		

CONCLUSION

Since the study was conducted in a controlled manner, the phytochemical results can be used for the standardization of the above mentioned drugs. A preliminary screening and more research has to be undertaken to explore the wonderful therapeutic properties of these medicines. To conclude the presence study, we have found that most of the biologically active phytochemicals were present in the ethanolic extract of the *Ficus racemosa* and *Erythrina variegata* leaves.

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