ORIGINAL RESEARCH

PRELIMINARY PHYTOCHEMICAL SCREENING OF *GARDENIA RESINIFERA* ROTH.

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ABSTRACT:

The Present study, deals with phytochemical screening of *Gardenia* species by using different types of solvent which shows slight variation in presence of chemicals or secondary metabolites these are noted in tabulation forms and compared with earlier records.

KEYWORDS: *Gardenia*, solvent, chemicals, and secondary metabolites.

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INTRODUCTION:

The subject of phytochemistry or plant chemistry has developed in recent years as a distinct discipline somewhere in between natural products, organic chemistry and plant biochemistry and closely related to both. It is concerned with the enormous variety of organic substances that are elaborated and accumulated by plants and deals with the chemical structure of these substances, their biosynthesis, turnover and metabolism, their natural distribution and their biological function[7].

In all these operations methods are needed for separation, purification and identification of the many different constituent present in plants-thus advances in our understanding of phytochemistry are directly related to the successful exploitation of known technique, and the continuing development of new technique to solve outstanding problem as they appears. One of the challenges of phytochemistry is to carry out all the above operation on vanishingly small amount of material frequently, the solution of a biological problem in say plant growth regulation in the biochemistry of plant animal interactions or in understanding the origin of fossil plant depends on identifying a range of complex chemical structure which may only be a available for study in micro gram amount. The range and number of discrete molecular structure produced by plant is huge and such the present rate of advance of knowledge of them that a major problem in phytochemical research is the collection of existing data on each particular class of compound. Phytochemical has been aided enormously by the development of rapid method and accurate of screening plants for particular chemical and the emphasis is inevitably on chromatographic technique. These procedure have shown that many substances originally thought to be rather are in occurrence are of almost universal distribution in the plant kingdom[7]. The importance of continuing surveys or plant for biologically active substance needs no stressing. As a general rule method used with higher plants for identifying alkaloids, amino-acid, quinines and terpenoids can be applied directly to microbial system. Phytochemical is natural bioactive compound found in plant such as vegetables, fruits, medicinal plants. Flowers, leaves and root that work with nutrients and fibers to acts as an defense system against disease or more accurately to protect against disease, phytochemical are divided into two groups *viz*., primary and secondary constituents, according to their function in plant metabolism. Primary constituents comprise common sugars, amino acid, proteins and chlorophyll and while secondary constituents consists of alkaloids, terpenoids and phenolic compound and many more such as flavonoids, tannins. In India the herbal drug market is about 1 billion and the export of plant based crude drugs is around 80 million[10]. But the most important challenges faced by these formulations arises because of their lack of complete standardization. Herbal medicines are prepared from materials of plant origin which are prone to contamination, deterioration and variation in composition. Therefore quality control of herbal medicine offers a host of problems. To solve this problem, first and foremost task is the selection of the right kind of plant material which is therapeutically efficacious. Natural product chemistry research is the backbone of herbal industry and directly or indirectly responsible for both failure and success of herbal drugs for promoting the use of herbals in modern medicine. Phytochemistry should be envisaged for 1) Isolation, purification and characterization of new phytochemicals 2) Use of newly isolated phytoconstituents as "lead" compound for the synthetic design of analogues with either improved therapeutic activity or reduced toxicity 3) Conservation of lead phytoconstituents into medicinally important drugs[12].

During the past decade, the traditional systems have gained importance in the field of medicine. The World Health Organization estimates that 4 million people 80% of the world population presently use herbal medicine for some aspect of primary health care. Herbal medicine is a major component in all indigenous people traditional medicine and common element in Ayurvedic, Homeopathic, Naturopathic, traditional oriental and Native American and Indian medicine. Many drugs

commonly used today are of herbal origin because of their safety quality and efficacy. Indeed about 25% of the prescription drugs dispends in the USA contain at least one active ingredient derived from plant material.[2] In Indian system of medicine the gum Dikamali is one of the important drugs. The drug is antispasmodic, antiseptic, anthelmintic and stimulant. It is used as a sedative externally on scalp with other oil. The gum Dikamali of Madras crude drug trade has been identified in out analytical laboratory as the gum of *Gardenia* species. *Gardenia resinifera* and *Gardenia latifolia* are related to Rubiaceae family which parades a long list of plants of medicinal importance. *G. resinifera* is shrub or small tree. It bears small leaves and secretes gummy matrix or sap at the detached end of ear and stem. This substance known as Dikamali or Cumbi-gum. Cumbi gum is antispasmodic, expectorant, diaphoretic, carminative, anthelmintic, relieves constipation, pain, treat worms. Gum is antimicrobial, anthelmintic used in skin diseases.According to Ayurveda it increases appetite, astringent to bowels, relieves pain of bronchitis, vomiting and constipation. Gum contains flavonoids, gardenins, wagonin derivatives, deMetangeretin, nevadensin, hexacosyl p-coumarate.

Medicinal Uses:

1) Resin warmed in coconut oil is applied on forehead, throat and taken orally acts as anthelmintic.
2) Resin of *Gardenia resinifera* warmed in castor oil is applied on wound and cuts.
3) Gum a spoonful of exudates mixed in powdered sugar and consumed daily to cure hepatic disorder.
4) It is astringent, increases bowels, relieve pain, bronchitis, vomiting and constipation.
5) Two to three teaspoon of stem exudates mixed in equal quantity of honey is mixed in a cup of coconut milk and the formation is administered orally a day up to 24-28 days in hepatopathy.

MATERIAL AND METHOD:

Plant material i.e. leaves of *Gardenia resinifera* was collected from campus of Government Vidarbha Institute of Science and Humanities, Amravati.

Extraction: Plant material was first dried under shade and then powdered. The air dried powder was extracted in Soxhlet's assembly with distilled water and ethanol. The extracts obtained in solvent were concentrated, distilling off the solvent and evaporated to dryness. Extract obtained in solvent concentrated, solidified and weighed. It's percentage was calculated in terms of dry weight of plant material. The colour of the extract was noted in sample (Table I).

Chemical Test:

The solvent free extract obtained as above was then subjected to qualitative test for the identification of various plant constituents from the sample[11].

1) Detection of Alkaloids:

The small portion of solvent water and ethanol extract was transferred in test tube and was stirred with a few drops dilute hydrochloric acid and filtered. The filtrate is tested carefully with various alkaloidal reagent such as Mayer's reagent (cream ppt), Wagner's reagent (reddish-brown ppt), Hager's reagent (yellow ppt).

2) Detection of Glycosides:

A small quantity of water and ethanol extract was dissolved in 5ml of distilled water and filtered. The filtrate is subjected to Borntrager's test for detection of glycosides.

3) Detection of Sugars:

The small quantity of water and ethanol extract was dissolved in 5ml of water and filtrate. The filtrate was subjected to Fehling test, Benedict's test for detection of sugar.

4) Detection of Saponin:

About 1ml of extract or water and ethanol extract was diluted separately with distilled water to 20 ml and shaken in graduate cylinder for 15 minutes. One cm layer of foam indicates the presence of saponin.

5) Detection of Amino Acid and Protein:

Small quantity of water and ethanol extract is dissolved in 10 ml of distilled water

and filtered through Whatman No. 1 filter paper and the filtrate was subjected to Biuret test for detection of protein.

Small quantity of hydrolyzed extract added two drops of Ninhydrin solution and was subjected to Ninhydrin test for amino acid.

6) Detection of Fixed Oil & Fats:

A small quantity of water and ethanol extract was passed separately between two filter paper. Oil stains on the paper indicated the presence of fixed oil.

7) Detection of Phenolic Compounds & Tannins:

Small quantity of alcoholic and aqueous extracts in water was tested for the presence of phenolic compound and tannin with dilute ferric chloride solution (5%), lead acetate (10%) and 1% gelatin solution.

8) Detection of Flavonoids:

Few amount of extract is treated with 10% ammonium hydroxide solution. Yellow fluorescence indicated the presence of flavonoids.

9) Detection of Gums and Mucilages:

About 10 ml of aqueous and ethanol extract was added to 25 ml absolute alcohol with constant stirring. The precipitate was dried in air it was examined for swelling property.[9]

OBSERVATIONS AND RESULT:

Nature and colour of aqueous extracts of *Gardenia resinifera* leaves is shown in table I. The average percentage yield of various extracts is shown in figure 1. Preliminary phytochemical screening has been done of leaves of *Gardenia resinifera* Roth. and the result are incorporated in table II.

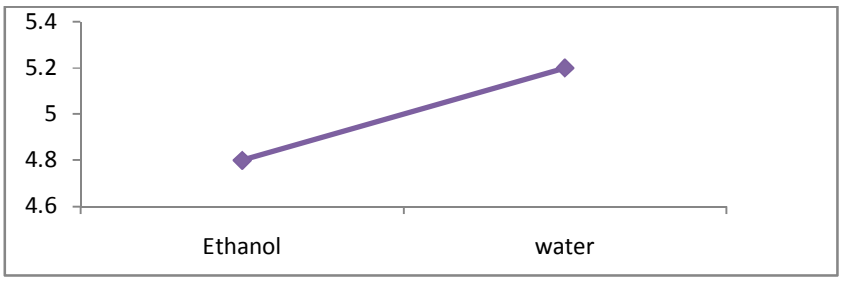
Table I: Nature and colour of various extracts of *Gardenia resinifera* Roth.

Extract	Texture	Colour
Distilled Water	Crystalline	Dark Brown
Ethanol	Crystalline	Green

Table II: Phytochemical Test:

Sr. No.	Chemical Constituent	Test	*Gardenia resinifera*	
			(Distilled Water)	Ethanol
1	Alkaloids	a) Mayer's test	–	++
		b) Wagner's test	+++	++
		c) Hager's test	+++	+
2	Sugar	a) Fehling test	+	+
		b) Benedicts test	–	+
3	Glycosides	a) Borntrager's test	–	++
4	Saponin		++	++
5	Protein and Amino acid	a) Biuret test	–	+
		b) Ninhydrin test	–	–
6	Fixed oils and Fats	a) Spot test		–
7	Phenolic compounds and Tannin	a) Ferric chloride	+++	+
		b) Gelatin test	–	–
		c) Lead acetate test	+++	+
8	Flavonoids	a) Alkaline reagent	–	+
9	Gum mucilage		+++	–

Fig.1 Percentage Extract of Crude Drug in Each Solvent of *Gardenia resinifera*.



High test for alkaloids in aqueous extract of leaves of *Gardenia resinifera* was observed with all three reagents. Aqueous extract showed high test Wagner's reagent and Hager's reagent and negative in Mayer's reagent. Ethanol extract showed high test in Mayer's test, Wagner's test and medium test in Hager's test.

Low test for sugar is found in aqueous extract with Fehling reagent, negative test in Benedict's test. In ethanol extract moderate test in fehling and Benedict's test.

Test for saponin was moderate in water and ethanol extracts. Phenolic compound and tannin were found to be present in high concentration with the water extract. Negative test for phenolic compound and tannin in water extract with Gelatin reagent and high test for ferric chloride and lead acetate test. In ethanol extract moderate test in ferric chloride and lead acetate test and negative in gelatin test.

High test for gum and mucilage is found in aqueous extract. In ethanol extract gum and mucilage test is negative.

Flavanoids test is moderate in ethanol extract and negative test in distilled water extract.Leaves extract of *Gardenia resinifera* have given negative test for glycosides, protein, amino acid, fixed oil and flavonoids in aqueous extract.While ethanol extract have negative test for amino acid, fixed oils and fats and gum and mucilage test.

DISCUSSION:

Anandkumar *et al.,* carried out the standardization of dikamali in *Gardenia resinifera* (Roth) and *Gardenia gummifera* Linn. They have found important oleoresin drug. In present investigation test for alkaloids, sugar, saponin, phenolic compound and tannin, gum mucilage was positive while glycosides, amino acid and protein, fixed oil and fats gave negative test in aqueous extract. In ethanol extract alkaloids, sugar, glycosides, saponin, protein, phenolic compound and tannin, flavanoids was positive while amino acid, fixed oil and fats were negative test[1].

Mai *et al*., have carried out chemosystematics of *Gardenia*. Test for different chemical constituent in 6 species of *Gardenia* is numerically analyzed. In present screening, test for alkaloids, sugar, saponin and gum were positive while it differs for the test of glycosides, amino acid, fats and oil and aqueous and ethanol extract [4].

Patwari *et al.,* have performed pharmacological screening of Dikamali and gum and resin isolated from *Gardenia resinifera* leaves. Phytochemical test showed presence of alkaloids, saponin and gum. In present investigation also test for these phytochemicals were positive in aqueous and ethanol extract[11].

Jhansi *et al*., carried out screening of secondary metabolites in methanolic leaf and bark extracts of *Gardenia resinifera* and *Gardenia latifolia*. In their study the extract showed presence of steroids, phenolic compound and flavonoids in leaf and bark. In present phytochemical screening extract gave positive test for alkaloids, saponin, gum mucilage and negative for glycosides, amino acid, protein, flavanoids in aqueous extract.

Ethanol extract gave positive test for alkaloids, sugar, glycosides, saponin, protein, phenolic and tannin and flavanoids and negative test for amino acid, fixed oils and fats and gum and mucilage[3].

Patwari *et al*., performed evolution of anti cancer activity of Dikamali. They have found different medicinal compound such as Dikamaliartane-A, a cycloartane were present. In present work phytochemical screening of leaves gave positive test for saponin, phenolic and tannin and alkaloids moreover test sugar were positive in aqueous and ethanol extract [6].

Salave, have carried out traditional hepatopathic treatment of medicinal plant from Newasa Tahsil of Ahmednagar district (M.S.), India. Traditional herbal therapies used in against various liver diseases [8].

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