ScienSage

Journal of Advanced Scientific Research

Available online through http://www.sciensage.info

ISSN
0976-9595
Short Communication

COMPARATIVE QUALITATIVE AND QUANTITATIVE PHYTOCHEMICAL SCREENING OF HYDROALCOHOLIC EXTRACT OF *GARDENIA LATIFOLIA* AND *GARDENIA RESINIFERA* LEAVES AND BARK EXTRACTS

Abhishek Patel*[1], Vishal Soni[2], Sarang Kumar Jain[3]

[1]*Research Scholar, B.R. Nahata College of Pharmacy, Faculty of Pharmacy, Mandsaur University, Mandsaur, Madhya Pradesh, India*

[2]*B.R. Nahata College of Pharmacy, Faculty of Pharmacy, Mandsaur University, Mandsaur, Madhya Pradesh, India*

[3]*Rajeev Gandhi College of Pharmacy, Bhopal, Madhya Pradesh, India*

**Corresponding author: abhipharma20@rediffmail.com*

ABSTRACT

The plant screening for phytochemical constituents seems to have the potential to act as a source of useful drugs and cures many infections as a result of the presence of various bioactive compounds that evident to have enormous activity against array human pathogens. The objective of the study was to undertake a comparative qualitative phytochemical analysis of the different parts like leaves and bark of *Gardenia latifolia* and *Gardenia resinifera* leaves, a traditional herb used against several diseases. The total flavonoid content in *Gardenia resinifera* bark and leaves was found 0.356 and 0.481 mg/100mg of dried extract. In comparison to *Gardenia resinifera,* the results of estimation of total phenol, flavonoids and alkaloid content of *Gardenia latifolia* were reported. The total phenolic content in hydroalcoholic extract of *Gardenia latifolia* bark and leaves was found 0.509 and 1.100 mg/100 mg of dried extract respectively. The total flavonoid content in *Gardenia latifolia* bark and leaves was found 0.325 and 0.556 mg/100 mg of dried extract. The total alkaloid content in *Gardenia latifolia* bark extract was found 0.351 mg/100 mg of dried extract. These findings suggested that *Gardenia latifolia* and *Gardenia resinifera* leaves and bark extract could be a potential source of drugs which in future may serve for the production of synthetically improved therapeutic agents.

Keywords: *Gardenia latifolia, Gardenia resinifera*, Phytochemical screening, Gallic acid, Quercetin.

1. INTRODUCTION

Over three-quarters of the world, population relies mainly on plants and plant extracts for health care and more than 30% of the entire plant species, at one time or the other was used for medicinal purposes [1]. The use of plants for medicinal purpose is probably as old as the history of mankind and their uses in the industrialized societies have led to the extraction and development of several drugs from as well as from traditionally used folk medicine. Extraction and characterization of several active phytochemicals from these green factories have given birth to some high activity profile drugs [2]. It has been estimated that in developed countries such as United States, plant-drived drugs constitute as much as 25% of the total drugs, while in fast developing countries such as China and India, the contribution is as much as 80%. Thus, the economic importance of medicinal plants is recognized in all countries over the world and these provide two third of the plants used in modern system of medicine and the health care system of rural population depend on indigenous systems of medicine [3]. Phytochemical studies have attracted the attention of plant scientists due to the development of new and sophisticated techniques. These techniques played a significant role in the search for additional resources of raw material for pharmaceutical industry (phyto-chemicals) [4]. Development of drugs based on natural products has had a long history in the US, and in 1991, almost half of the best selling drugs were natural products or derivatives of natural products [5]. Natural products are chemical compounds derived from living plants or animals. Drugs derived from natural products are usually secondary metabolites and their derivatives Comparison of the phytochemical composition of different plant parts may lead to the utilization of plants parts, in particular the aerial parts, with minimum adversity to the conservation of the plants.

Gardenia latifolia, commonly known as Indian boxwood or Ceylon boxwood, is a small tree with dense foliage. The different parts of this plant are reported to be used in

treatment of a wide range of ailments such as snake bite, skin diseases, stomach pains, inflammatory pain, caries, haemorrhage in humans and ephemeral fever in live stocks [6-8].

Gardenia resinifera is native to both tropical and subtropical regions of Asia, Africa1 etc. [9], it is a flowering plants in the coffee family, Rubiaceae. *Gardenia resinifera* are evergreen shrubs and small trees growing to 1-15 m (3.3-49.2 ft) tall. Flowering is from mid-spring-midsummer. Gardenia is commonly used to treat infections, particularly bladder infections, abscesses, jaundice and blood in the urine, sputum, or stool. It is also used to treat anxiety or insomnia. It is also helpful in menopausal imbalance reflected in insomnia and depression, nervous tension, dizziness, and headache. This plant is rich in diverse chemical constituents like methyl 7-keto-octadec-cis-ll-enoic acid [10], dikamaliartanes A-F [11], Gardenin A, B, D, E, 5-Desmethylnobiletin, Xanthomicrol and Acerosin [12]. Some special pharmacological activities reported on *Gardenia resinifera* were antispasmodic, expectorant, anti-microbial and anti- helmintic [13], Anti-epileptic, anti-pyretic [14] and anti- convulsant activities [15], anti-proliferative activity against lung [16], breast, colon, hepatic and leukemia cell lines. Due to its broad spectrum healing potential, this medicinal tree exhibits itself as a very good research material for various scientific studies. In this regard, the primary aim of the study was to undertake the qualitative and quantitative phytochemical analysis of different parts (leaves and roots) of *Gardenia latifolia* and *Gardenia resinifera* with intention of motivation for usage of plant parts with less adverse implications for the survival of the plant species.

2. MATERIAL AND METHODS

2.1. Plant material

The plant *Gardenia latifolia* and *Gardenia Resinifera* (leaves and bark) were collected from local area of Bhopal (M.P.) in the month of July, 2020. The leaves and bark plant sample were separated and washed with sterile distilled water to remove the adhering dust particles and other unwanted materials. The leaf and bark were air dried under room temperature. The dried plant samples were cut and grinded to make it in powder form.The powdered samples were stored in clean, dry and sterile container for further use.

2.2. Chemical reagents

All the chemicals used in this study were obtained from Hi Media Laboratories Pvt. Ltd. Mumbai, India), SD Fine- Chem. Ltd. (Mumbai, India) and SRL Pvt. Ltd. (Mumbai, India). All the chemicals used in this study were of analytical grade. Quercetin and gallic acid was kindly provided by Scan Research Laboratories, Bhopal (India).

2.3. Extraction

The extraction of bioactive secondary metabolites from plant materials is an important phase in phytochemical processing. The choice of an appropriate extraction technique is also essential for the standardization of herbal products. With the aid of chosen solvents, extraction is used to remove suitable soluble constituents while excluding those that aren't. The plant materials were vigorously rinsed in purified water after being thoroughly washed in tap water. The washed, stable plant samples were cut into small pieces and dried for 3 to 4 weeks in the shade.

The dried plant samples of *Gardenia resinifera* and *Gardenia latifolia* were ground into fine powder using an electric grinder after the drying process. The powdered plant samples were then held in the fridge and used for extraction. For the preparation of ethanol and aqueous extracts from shade dry samples of various sections of selected plants, the following extraction technique was used. For small research projects, maceration extraction is a popular technique [17-18].

2.4. Defatting of Plant Material

A 120 gram bark, 110 gram leaves shade dried powder of *Gardenia resinifera* and 100 gram bark, 60 gram leaves shade dried powder of *Gardenia latifolia* were extraction with petroleum ether using Soxhlet extraction process. The extraction was continued till the defatting of the material had taken place.

2.5. Successive extraction with different solvents by maceration method

Defatted plant material of *Gardenia resinifera* and *Gardenia latifolia* were extracted with ethanol: water (70:30) by Soxhlet extraction process [17]. The resultant content was filtered with whatman filter paper no.1 and kept for evaporation of solvent to get the dry concentrated extract. The dried crude concentrated extract was weighed to calculate the extractive yield then transferred to glass vials (6×2 cm) and stored in a refrigerator (4°C), till used for analysis.

2.6. Determination of extractive value (% yield)

The extraction yield is evaluate of the solvent's efficiency to extracts bioactive components from the selected

natural plant samples and it was defined as quantity of plant extracts recovered in mass after solvent extraction compared with the initial quantity of plant samples. After extraction, yield of the plant extracts obtained were calculated in grams and then converted it into percentage. Following formula was adopted for determination of percentage yield of selected plant materials.

2.7. Calculation of % yield

The % yield of yield of each extract was calculated by using formula:

Percentage yield = (Weight of extract/Weight of powered drug taken) x 100

2.8. Qualitative Phytochemical analysis

Medicinal plants are resources of traditional medicines and many of the modern medicines are produced indirectly from plants. Phytochemical constituents are of two type primary bioactive constituents (chlorophyll, proteins, amino acids, sugar etc.) and secondary bioactive constituents include (alkaloids, terpenoids, phenols, flavonoids etc.)

Phytochemical screening is a process in which the extraction, identification and screening of phytochemicals can be done easily for variety of medicinal plants. As these phytochemicals are precursors for the preparation of various new drugs. Hence, the plant extracts obtained by aqueous and ethanol extraction of all selected plant samples were subjected to various qualitative screening to detect the presence of plant bioactive constituents present in them. Preliminary phytochemical screening is primarily an important aspect for establishing profile of given extract for its chemical compounds produced by plant. Phytochemical examinations were carried out extracts as per the following standard methods.

2.9. Methods of Phytochemical screening

The methods of Khandelwal, (2005), Kokate, (1994) [17-18] were adopted for the preliminary phytochemicals screening. Following phytochemical tests were used for qualitative screening of all the plant samples extracts to examine the presence of alkaloids, carbohydrates, glycosides, flavonoids, phenols, saponins, tannins, terpenoids, and proteins and amino acids [18, 19].

2.10. Quantitative studies of phytoconstituents

2.10.1. Estimation of total phenol content

The total phenol content of the extract was determined by the modified folin-ciocalteu method [20-22]. 10 mg Gallic acid was dissolved in 10 ml methanol, various aliquots of 10-50µg/ml was prepared in methanol. 10 mg of dried extract was dissolved in 10 ml methanol and filter. Two ml (1mg/ml) of this extract was for the estimation of phenol. 2 ml of extract and each standard was mixed with 1 ml of Folin-Ciocalteu reagent (previously diluted with distilled water 1:10 v/v) and 1 ml (7.5g/l) of sodium carbonate. The mixture was vortexed for 15s and allowed to stand for 10min for colour development. The absorbance was measured at 765 nm using a spectrophotometer.

2.10.2. Estimation of total flavonoids content

Determination of total flavonoids content was based on aluminium chloride method [23]. 10 mg quercetin was dissolved in 10 ml methanol, and various aliquots of 5-25µg/ml were prepared in methanol. 10mg of dried extract was dissolved in 10 ml methanol and filter. Three ml (1mg/ml) of this extract was for the estimation of flavonoids.1 ml of 2% $AlCl_3$ solution was added to 3 ml of extract or each standard and allowed to stand for 15min at room temperature; absorbance was measured at 420 nm.

2.10.3. Estimation of total alkaloids content

The plant extracts (1mg) was dissolved in methanol, added 1ml of 2 N HCl and filtered. The solution was transferred to a separating funnel, 5 ml of bromocresol green solution and 5 ml of phosphate buffer were added. The mixture was shaken with 1, 2, 3 and 4 ml chloroform by vigorous shaking and collected in a 10-ml volumetric flask and diluted to the volume with chloroform. A set of reference standard solutions of atropine (40, 60, 80, 100 and 120µg/ml) were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the reagent blank at 470 nm with an UV/Visible spectrophotometer. The total alkaloid content was expressed as mg of AE/100mg of extract [10].

3. RESULTS AND DISCUSSIONS

Bark and leaves of *Gardenia resinifera* and *Gardenia latifolia* were extracted using hydroalcoholic solvent. Extractive values are mainly used to determine whether a substance is exhausted or adulterated, and they are a valuable method for determining the drug's consistency and difference in chemical constituents. The extractive values are measures of the overall soluble component in the solvent. Results of extractive value showed in table 1.

Phytochemical constituents such as phenol, flavonoids, alkaloids or secondary metabolites possess nutritive and

pharmacological activities. Thus the preliminary screening test may be useful in the detection of the bioactive principles and subsequently may lead to the drug discovery and development. The results of qualitative phytochemical analysis of *Gardenia resinifera* (leaves and bark) extracts were presented in table 2. The results revealed the presence of flavonoids, phenol, diterpenes, proteins, carbohydrate and saponins in leave and bark extract. There was absence of alkaloids and glycosides in leave and bark extract of *Gardenia resinifera*. The results of phytochemical investigation of *Gardenia latifolia* were presented in table 3. The results revealed the presence of flavonoids, phenol, proteins, carbohydrate and saponins in bark extract. There was absence of alkaloids, glycosides and diterpenes in bark extract of *Gardenia latifolia*. The results of leaves extract of *Gardenia latifolia* showed the presence of alkaloids, flavonoids, diterpenes, phenol, proteins, carbohydrate and saponins. Only glycoside was found to be absent in hydroalcoholic leaves extract of *Gardenia latifolia* (table 3).

Total phenolic compounds (TPC) was expressed as mg/ 100mg of gallic acid equivalent of dry extract sample using the equation obtained from the calibration curve: Y = 0.011X+0.011, R^2= 0.998, where X is the gallic acid equivalent (GAE) and Y is the absorbance. Total flavonoids content was calculated as quercetin equivalent (mg/100mg) using the equation based on the calibration curve: Y=0.032X + 0.018, R^2=0.998, where X is the quercetin equivalent (QE) and Y is the absorbance. Total alkaloid content was calculated as atropine equivalent mg/100mg using the equation based on the calibration curve: Y=0.007X+ 0.024, R^2=0.995, where X is the Atropine equivalent (AE) and Y is the absorbance. The results of total phenols, flavonoid and alkaloids were presented in table 4 & 5. The total phenolic content in hydroalcoholic extract of *Gardenia resinifera* bark and leaves was found 0.660 and 0.600mg/100 mg of dried extract respectively. The total Flavonoid content in *Gardenia resinifera* bark and leaves was found 0.356 and 0.481 mg/100 mg of dried extract. In comparison to *Gardenia resinifera* the results of estimation of total phenol, flavonoids and alkaloid content of *Gardenia latifolia* were represent in table 5. The total phenolic content in hydroalcoholic extract of *Gardenia latifolia* bark and leaves was found 0.509 and 1.100 mg/100 mg of dried extract respectively. The total flavonoid content in *Gardenia latifolia* bark and leaves was found 0.325 and 0.556 mg/100 mg of dried extract. The total alkaloid content in *Gardenia latifolia* bark extract was found 0.351 mg/ 100 mg of dried extract.

Table 1: Extractive values of Hydroalcoholic extract of *Gardenia resinifera and Gardenia latifolia*

S. No.	Plants	Plant Parts	% Yield (W/W)
1.	*Gardenia resinifera*	Bark	5.90
		Leaves	4.55
2.	*Gardenia latifolia*	Bark	7.46
		Leaves	6.21

Table 2: Result of Phytochemical screening of bark extract of *Gardenia resinifera*

S. No.	Constituents	Hydroalcoholic bark extract	Hydroalcoholic leaves extract
1.	**Alkaloids**		
	Hager's Test:	-ve	-ve
2.	**Glycosides**		
	Legal's Test:	- ve	- ve
3.	**Flavonoids**		
	Alkaline Reagent Test:	+ ve	+ ve
	Lead acetate Test:	+ ve	- ve
4.	**Diterpenes**		
	Copper acetate Test:	+ ve	- ve
5.	**Phenol**		
	Ferric Chloride Test:	+ve	+ve
6.	**Proteins**		
	Xanthoproteic Test:	+ve	+ ve
7.	**Carbohydrate**		
	Fehling's Test:	+ ve	+ ve
8.	**Saponins**		
	Froth Test:	+ve	+ ve

Table 3: Result of Phytochemical screening of hydroalcoholic extract of *Gardenia resinifera*

S. No.	Constituents	Hydroalcoholic bark extract	Hydroalcoholic leaves extract
1.	**Alkaloids**		
	Hager's Test:	-ve	+ve
2.	**Glycosides**		
	Legal's Test:	- ve	- ve
3.	**Flavonoids**		
	Alkaline Reagent Test:	+ ve	+ ve
	Lead acetate Test:	- ve	- ve
4.	**Diterpenes**		
	Copper acetate Test:	- ve	+ ve
5.	**Phenol**		
	Ferric Chloride Test:	+ve	+ ve
6.	**Proteins**		
	Xanthoproteic Test:	+ ve	+ve
7.	**Carbohydrate**		
	Fehling's Test:	+ ve	+ ve
8.	**Saponins**		
	Froth Test:	+ ve	+ve

Table 4: Estimation of total phenol, flavonoids and alkaloid content of *Gardenia resinifera*

S. No.	Hydroalcoholic Extract	Total phenol content	Total flavonoids content	Total alkaloid content
			(mg/ 100 mg of dried extract)	
1	Bark	0.660	0.356	-
2	Leaves	0.600	0.481	-

Table 5: Estimation of total phenol, flavonoids and alkaloid content of *Gardenia latifolia*

S. No.	Hydroalcoholic Extract	Total phenol content	Total flavonoids content	Total alkaloid content
			(mg/ 100 mg of dried extract)	
1	Bark	0.509	0.325	0.351
2	Leaves	1.100	0.556	-

4. CONCLUSION

Qualitative and quantitative phytochemical analysis of plant parts extracts is important as it indicate the nature of phytochemicals that are possessed by such medicinal plants. The results of the current study suggest more similarities in the phytochemical compositions of the different parts of *Gardenia resinifera* and *Gardenia latifolia* which is likely to contribute to some similarities in their biological activities.

5. REFERENCES

1. Akroum S, Satta D, Lalaoui K. *Eur. J. Sci. Res*, 2009; **2:**289-295.
2. Sato OY, Singyouchi H, Ohtsubo K, Kihara T, Shibata HM. *International Journal of Pharmaceutical Sciences and Research*, 1997; **20:**401-403.
3. Joy PP, Thomas J, Samuel M, Baby PS. *Aromatic and medicinal plants*, Kerela agricultural University, India, 1998, Pp 44-46.
4. Mongole AJ, Awati R, Chaturvedi A, Zanwar P. *International Journal of Pharm Tech Research*, 2010; **2:**2307-2312.
5. Sneden AT. Natural Products as Medicinally Useful Agents, 2004; Pp21-23.
6. Vindhya K, Sampath KKK, Neelambika HS, Leelavathi S. *Research Journal of Pharmaceutical, Biological and Chemical Sciences,* 2014; **5(2):**527-532.
7. Sinha K, Mishra NP, Singh J, Khanuja SP. *Indian J Trad Know* 2004; **3:**257-270.
8. Vigneswaran M, Nanthakumaran T, Shanthasubitha S, Shanmugapriya B. *Indo-Asian Journal of Multi-disciplinary Research,* 2016; **2(1):**494-505.

9. Tao C, Charlotte M. Taylor, "Gardenia J. Ellis, Philos. Trans. 51: 935. 1761", Flora of China (FOC) Vol 19 pg.57,8,59,65 141
10. Chirag M, Daulatabad JD, Mulla GMM, Mirajkar AM, Hosamani KM. *Phytochemistry,* 1991; **30:**2399-2400.
11. Olaf K, Gandhe S, Gummadi SB, Belvotagi VR, Adavi R, Marupaka R, et al. *Chemistry & Biodiversity,* 2009; **6:**1185-1192.
12. Priyanka M, Shilpi S, Madan MG, Suaib L. *Biomed. Pharmacothera*., 2016; **85:**444-456.
13. Jhansi LB, Jaganmohanreddy K. *Biosci. Biotech. Res. Comm,* 2011; **4:**23-28.
14. Sridhar PG, Rama NRA, Appa Rao AVN, Narsimha Reddy Y. *Dikamali,* 2011; **2:**45-80.
15. Sridhar SK, Ramachandran S, Anbalagan N, Leonard JT, Joanofarc JKS. *Natural Product Sciences,* 2003; **9(1):**10-12.
16. Jhansi Lakshmi B, Jaganmohanreddy K. *Biosciences Biotechnology Research Asia*, 2013; **10(1):**275-281.
17. Khandelwal KR. Ed. Practical Pharmacognosy Technique and Experiments, 23rd Edn: 2005; 15.
18. Kokate CK. Ed. Practical Pharmacognosy, 4th Edn., *Vallabh Prakashan*: 1994; 112:120.
19. Mukherjee PK. Quality Control of Herbal Drugs, 2nd Edition, Business Horizons, 2007; 2-14.
20. Roopashree TS, Dang R, Rani SRH, Narendra C. *International Journal of Applied Research in Natural Products,* 2008; **1(3):**20-28.
21. Obasi NL, Egbuonu ACC, Ukoha PO, Ejikeme PM. *African Journal of Pure and Applied Chemistry,* 2010; **4(9):**206-212.
22. Audu SA, Mohammed I, Kaita HA. *Life Science Journal,* 2007; **4(4):**7579.
23. Olajuyigbe OO, Afolayan AJ. *BMC Complement Altern Med,* 2011; **11:**130.
24. Rohit S, Hetal A, Prajapati PK. *The Journal of Phytopharmacology,* 2015; **4(2):**116-120.