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Research Article

Comparative Phytochemical Studies and Evaluation of Radical Scavenging Activity in Selected *Jasminum* Species

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ABSTRACT
Phytochemical studies were carried out in five Jsaminum species. The chemical profiles of different species were compared using chromatographic techniques such as TLC and HPLC. The total phenolic content (TPC) and radical scavenging capacity were also evaluated. The DPPH EC_{50} value was found least for *Jasminum Grandiflorum* (7.5µg) showing its highest antioxidant activity. The EC $_{50}$ (DPPH) values vary as, 15 µg *(J.angustifolium*), 12 µg *(J.auriculatum*), and 7.5µg (*J. grandiflorum).*

Key words: TLC, HPLC, DPPH, Total phenolics.

INTRODUCTION

Medicinal plants and plant- derived medicines are widely used in traditional cultures all over the world and they are becoming increasingly popular in modern society as natural alternatives to synthetic chemical (Van Wyk and Michael Wink, 2009*)*. Nearly all cultures from ancient times have used plants as a source of medicine. The World Health Organization (WHO) has listed 21,000 plants worldwide, reported to have medicinal uses. India is the largest producer of medicinal herbs and is called the *botanical garden of the world* (Trivedi, 2007).

Jasminum genus with about 200 species belonging to family Oleaceae are of three types: shrub or bush form, vines and trees, native to tropical and warm temperate regions. Many jasminum plants prominently feature white, yellow or pink flowers with sweet fragrance and others are unscented Jasminum species is used to treat many conditions such as amenorrhoea, leprosy, skin diseases and also as an analgesic, antidepressant and anti-inflammatory (http://www.ehow.com/about_6325794_jasmine-plants).

Jasminum grandiflorum is a scrambling sub erect twining evergreen shrub (Anonymous, 2004), native to India, France, Italy, China, Japan, Morocco and Egypt [Chopra *et al*, 1958].The leaves are opposite, entire ovate to somewhat elliptic in shape with acuminate mucronate apex, whereas flowers are terminal and axillary cymes, calyx lobes are long, linear (Cooke, 1967 & Nadkarni, 1976). Roots are useful in cephalalgia, mental debility, chronic constipation, flatulence, strangury, sterility, dysmenorrhoea, amenorrhoea, ringworm, leprosy, skin diseases and giddiness.

Jasminum sambac is commercially grown in India, Thailand, China and Philippines. It is an evergreen vine or shrub reaching up to 1-3 m. The leaves are ovate; phyllotaxy is opposite or in whorls of three. The flowers blooms throughout the year and are produced in clusters of 3-12 together. The plant traditionally used as an analgesic, antidepressant, anti inflammatory, antiseptic, aphrodisiac, sedative, expectorant and tonic (uterine) effects. Roots are used to treat wounds and snake bites. The leaves and flowers have antipyretic and decongestant properties. Phytochemical studies shown that the roots contains dotriacontanoic acid, dotriacontanol, oleanolic acid, daucosterol and hesperidin (Zhang *et al*, 2004) and leaves contain sambacosides A, E and F (Tanahashi, 1988) flower contains molihuaside A-E, sambaeoside A (Zhang *et al*, 1995).

Jasminum angustifolium, distributed in south India (kerala, Karnataka) on the hills of lower elevation (Bown, 1995) Leaves are simple ovate-lanceolate, acute, glabrous and flowers are either solitary or more usually in three. Petals are linear, obtuse and acute Hepatoprotective effect of ethanolic and chloroform extract of *Jasminum angustifolium* were evaluated against carbon tetrachloride (1ml/kg) induced hepatic damage and was evidenced by reduction in level of alkaline phosphatase (ALP), alkanine amino transferase (ALT), aspartate amino transferase (AST), cholesterol, glucose, total protein and bilirubin concentration in blood (Joshi *et al*, 2008).

Jasminum auriculatum grows almost throughout South India, on dry slopes of the Western Ghats. The roots are useful in skin diseases especially for ringworm and flowers are fragrant, bitter, acrid, sweet, refrigerant, astringent, cardiotonic, diuretic and depurative in nature. They are useful in burning sensation, hyperdesia, ulcers, odontalgia, stomatopathy, ophthalmopathy, cardiopathy, urolithiasis, nephrolithiasis, strangury and dermatopathy (Ghosh, 1984). *Jasminum auriculatum* leaves have been reported to contain lupeol and jasminol (Deshpande *et al*, 1967). Alcoholic and aqueous extracts of flowers of *Jasminum auriculatum* showed diuretic activity by increasing the total volume of urine and concentrations of potassium and sodium salts in urine and antiurolithiatic

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Fig: 1.1 TLC (UV 254)

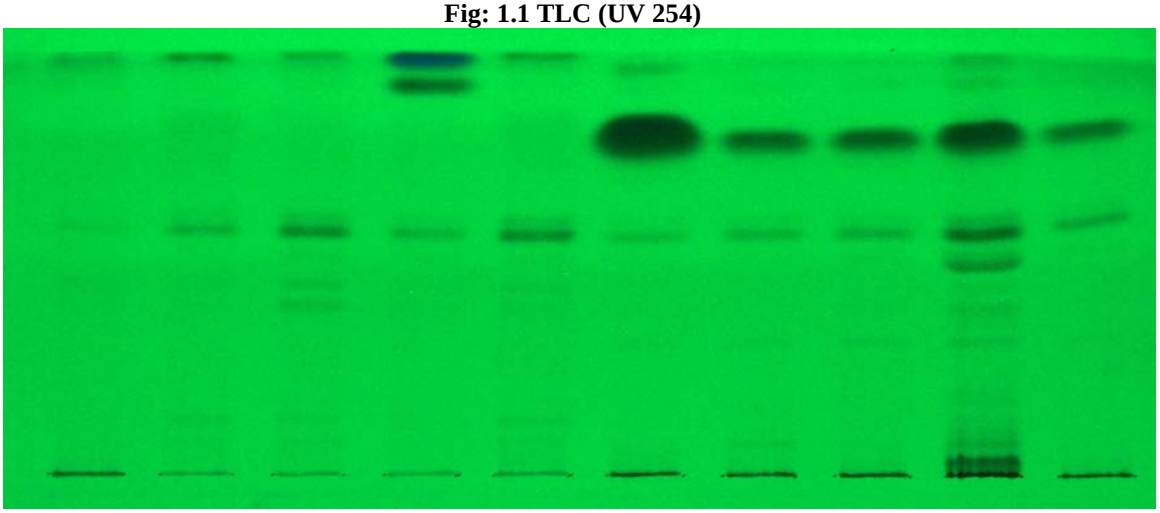


Fig: 1.1 TLC (UV 366 nm)

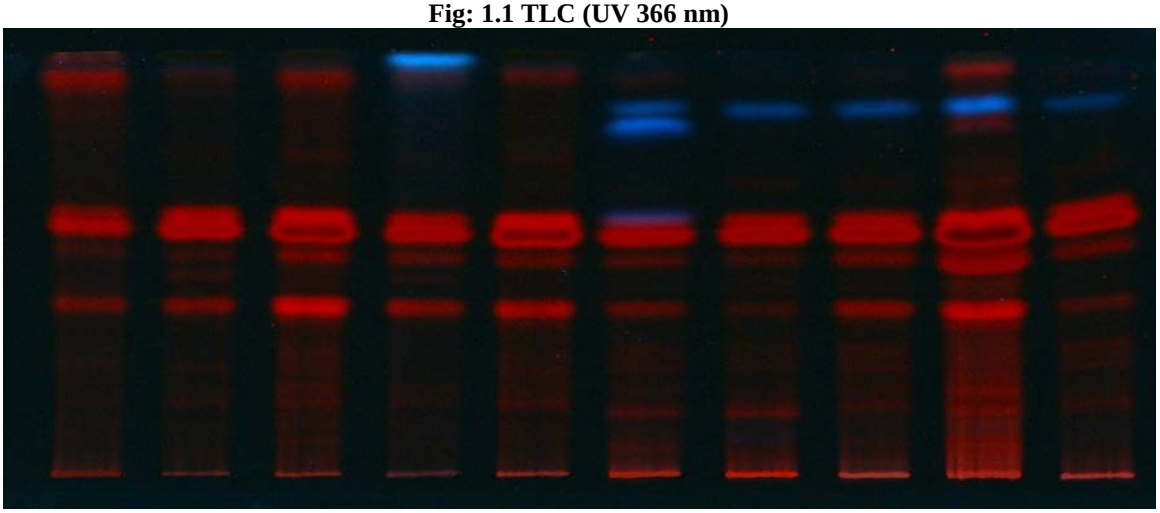
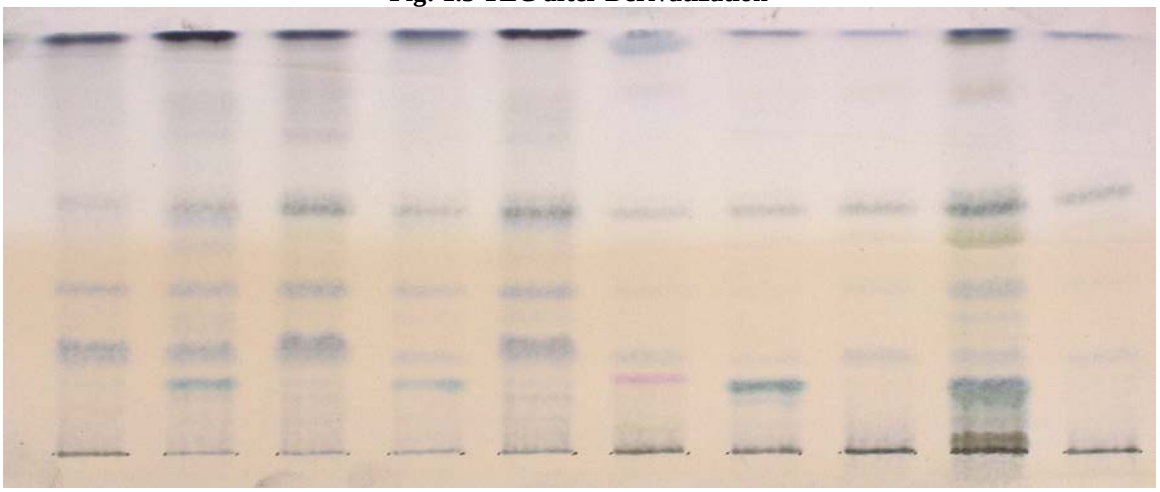


Fig: 1.3 TLC after Derivatization



activity by reducing the elevated urinary oxalate synthesis. (Bahuguna *et al*, 2009).

MATERIALS AND METHODS

Extraction: Plant materials (leaf) were collected from Herb Garden, Arya Vaidya Sala- Kottakkal and authenticated by Taxonomy division of Centre for Medicinal Plant Research, Arya Vaidya Sala, Kottakkal, Kerala. The powdered samples (2g each) were subjected to sequential extraction using 100ml of petroleum ether, chloroform, and methanol respectively for 5 hours. The final volume is made up to 50 ml to get 40-mg/ ml solution. These solutions are taken for different analyses.

TLC Analysis: TLC analysis of petroleum ether and chloroform extracts was carried out on a precoated silica plate (F_{254} Merck) using toluene: ethyl acetate as mobile phase in the ratio 9:1. TLC analysis of methanol extracts was carried out using toluene: ethyl acetate as mobile

Fig: 1.4 TLC of Methanol Extracts

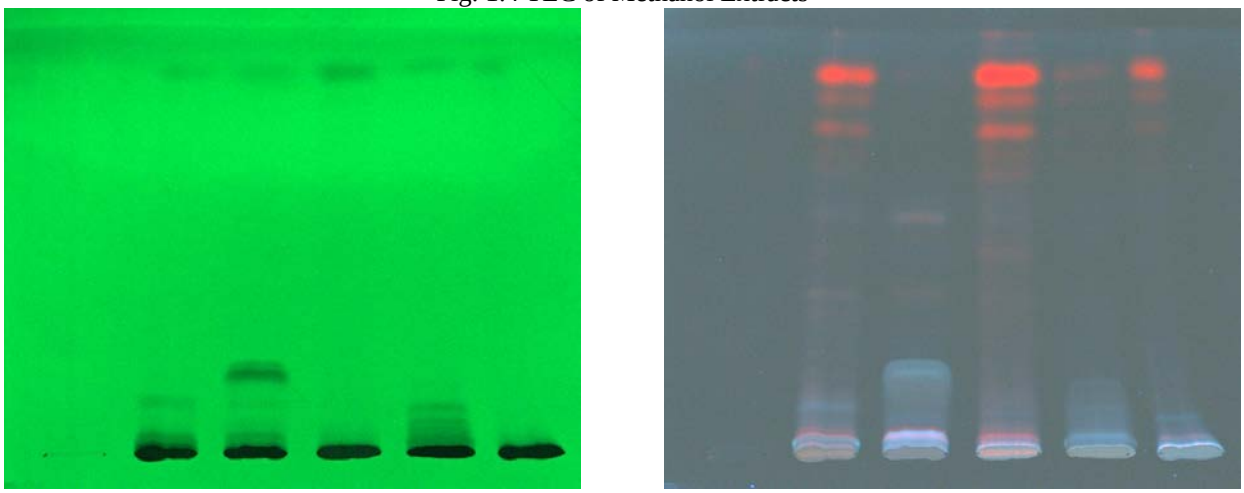
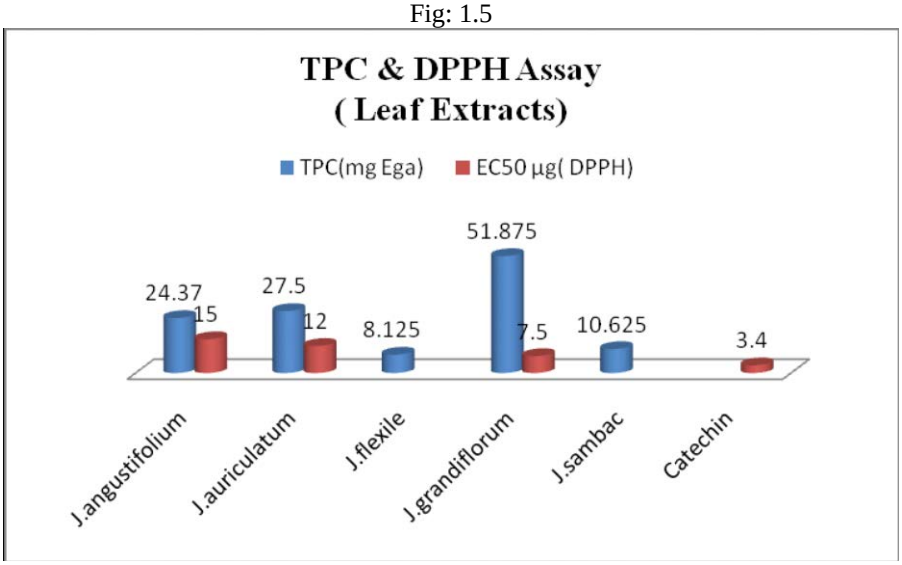


Fig: 1.5



phase in the ratio 8:2. The plates were developed up to 9cm and visualized under UV 254 nm, UV 366 nm and in visible light after derivetizing with Anisaldehyde-sulphuric acid (ANS) reagent.

HPLC Analysis: HPLC profiling was done using a Shimadzu High Performance Liquid Chromatographic system equipped with LC-10ATVP pump, SPD M10AVP Photo Diode Array Detector in combination with CLASS-VP 6.12 SP5 integration software. The mobile phase used for the separation was HPLC grade 0.1% formic acid in Acetonitrile (A) and methanol (B) in a time programming 0-10 10% A, 10-20 30% A, 20-30 50% A, 30-40 60% A and 40- 50 70 % A. . The column used was C18 – ODS (Octadecylsilane), Lichrospher RP 18e (5µm) (Merck) with a Phenomenex guard column (4mm x 2 mm i.d: 5µm). The samples were injected using a 20 µl loop (Rheodyne Rohnet Park, CA, USA). The flow rate was maintained to 0.75 ml/min.

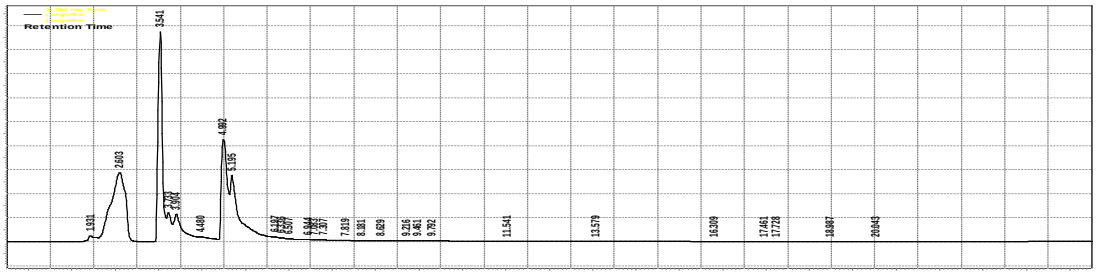
Total Phenolic Content (TPC): The assay was based on the reduction of phosphomolybdate ion of Folin-Ciocalteu reagent by the phenolate ion of sample. A desired amount of plant extract, distilled water and 1 N Folin-Ciocalteu reagent was taken into a tube and mixed thoroughly. After an interval of 3 min, 2 ml of 2% sodium carbonate solution was added and the mixture was allowed to stand for 30 min with intermittent shaking. The absorbance of the mixture was measured at 550 nm using spectrophotometer (Shimadzu, Japan). Different Gallic acid standards (2, 5, 7, 10, and 15 µg/ml) were used for obtaining a standard curve (Singleton *et a,l* 1965). The total phenolic content was expressed as Gallic acid equivalents (GAE) per gram of sample.

DPPH Assay: DPPH radical scavenging assay DPPH radical scavenging activity of the leaf extracts were determined according to the method described by Kukic *et al.* . First, 4.0 mL of test material at different concentrations were reacted with 0.50 mL of 1.0 mM DPPH solution and kept in the dark for 30 minutes, following which the absorbance was measured at 517 nm against a blank sample consisting of 4.0 mL of MeOH and 0.50 mL DPPH solution. DPPH scavenging activity was calculated using the equation:

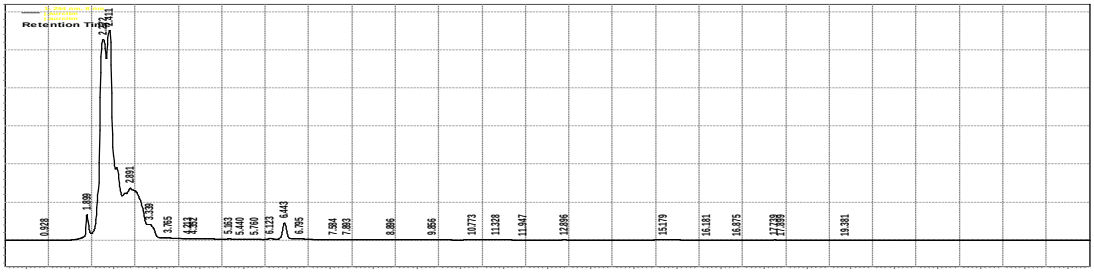
% DPPH radical scavenging activity = $(A_o-A_s/A_o) \times 100$.

Where A_o is the absorbance of the blank sample, and A_s is the absorbance of the test material. The IC_{50} value represented the concentration of test material that caused

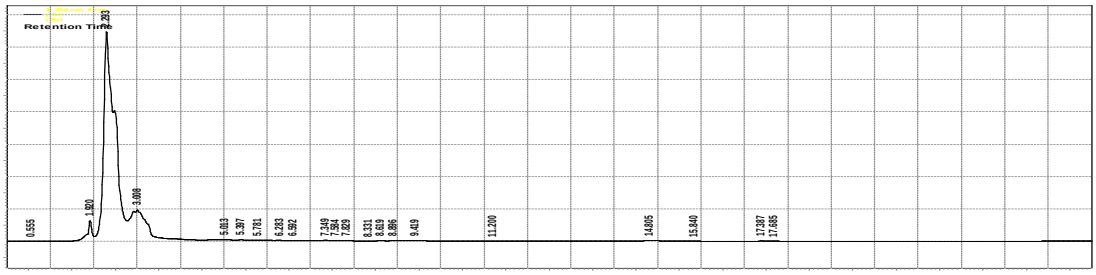
HPLC Profile of *Jasminum Angustifolium*



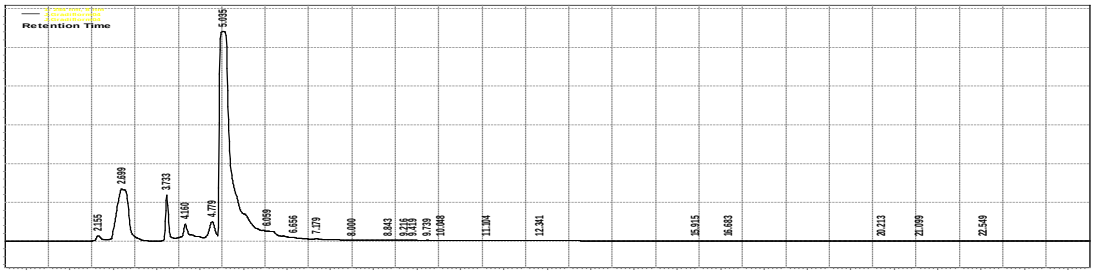
HPLC Profile of *Jasminum Auriculatum*



HPLC Profile of *Jasminum Flexile*



HPLC Profile of *Jasminum Grandiflorum*



50% scavenging activity. Catechin was used as positive controls.

RESULTS AND DISCUSSION

TLC Analysis: The thin layer chromatographic profiles of different extracts revealed the chemical pattern of each extract. On visualizing under 254 nm a compound with R_f 0.54 was found in both petroleum ether and chloroform extracts of all the species. A band with R_f 0.37 is present in petroleum ether extracts of *J.flexile, J. grandiflorum and J.sambac*, while the same is absent in *J.angustifolium* and *J. auriculatum*. The band with R_f 0.12 is present in the petroleum ether extracts of *J.auriculatm, J. flexile and J. sambac*. At R_f 0.85 an intense band is seen in the petroleum ether extract of *J. grandiflorm*. This particular compound is specific for *J. grandiflorum* (Fig: 1.1)

On visualizing under 366 nm compounds with Rf 0.37, 0.49, 0.55 are present in all species of petroleum ether extracts and chloroform extracts. A compound with Rf 0.91 is specific for the petroleum ether extract of *J. grandiflorum*. A compound with Rf 0.87 is present in petroleum ether extracts of *Jasminum Angustifolium, J. flexile,* and *J. sambac*.(Fig:1.2) For chloroform extracts, a common band was observed at R_f 0.80. The compounds with R_f 0.58, and 0.77 are specific for *J. angustifolium* (Figure 1.2).

After derivatisation with Anisaldehyde –Sulphuric acid (ANS) a compound with R_f 0.55 was present in both

petroleum ether and chloroform extracts of all the species. A compound at R_f 0.15 is found in both the petroleum ether extract and chloroform extract of *J. auriculatum* and *J. grandiflorum*. In the case of petroleum ether extracts a band at R_f 0.23 is found in all species except *J. grandiflorum* and a band at R_f 0.69 is found in *J. auriculatum, J. flexile* and *J. sambac*. A band with R_f 0.36 is present in all species of petroleum extracts (Figure 1.3).

For methanol extracts, on visualizing under 254 nm, it was found that a compound with R_f 0.83 is present in all species. At R_f 0.11 a compound is present in *J. angustifolium* and *J.grandiflorum* and at R_f 0.17 a compound is present only in *J. auriculatum* (Figure 1.4) On visualizing under 366 nm a pink colour band is present with R_f 0.04 in all species. A band at R_f 0.10 is found in *J. angustifolium, J. grandiflorum* and *J. sambac*. A compound with R_f 0.18 is present only in *J. auriculatum*. Compounds with R_f 0.71, 0.78 and 0.83 are found in all species except *J. auriculatum* (Figure 1.4).

Total Phenolic Content (TPC): A comparative determination of Total Phenolic Content (TPC) of methanol extracts of leaves of the five species was determined by Folin-ciocalteu phenol reagent using standard Gallic acid. The amounts of total phenolic content were expressed as mg Gallic acid Equivalents (GAE) per gram. It was found that the phenolic content is highest in *Jasminum Grandiflorum* (51.875mg GAE). The phenolics content of *J. flexile* (8.125 mg GAE) and *J. sambac* (10.625 mg GAE) are comparatively less (Figure 1.6)

DPPH Assay: The DPPH EC_{50} value was found least for *Jasminum Grandiflorum* (7.5µg) showing its highest antioxidant activity. The EC_{50} (DPPH) values vary as, 15 µg *(J.angustifolium*), 12 µg *(J.auriculatum*), and 7.5µg (*J. grandiflorum).* It was found that a positive correlation between total phenolic content and radical scavenging activity. *J. flexile* & *J. sambac* do not have radical scavenging activity.

HPLC Analysis: HPLC Analysis of methanol extracts was done for the comparison of their chemical profiles. In the chromatogram of *J. angustifolium* major peaks were found at R_t 2.603, 3.541, 3.904, 4.992 and 5.195. *J. auriculatum* showed major peaks at R_t 1.899, 2.272, 2.411, 2.891 and 6.443. The peaks at R_t 1.920, 2.293 and 3.008 are the major peaks in *J. flexile*. For *J. grandiflorum*, the major peaks obtained at R_t 2.155, 2.699, 3.733,4.160,4.779 and 5.035. The chromatogram of *J. sambac* showed peaks at R_t 1.984, 2.731, 3.723, 4.181 and 5.035.

The compound corresponds to R_t 1.9 was found in all species except *J. grandiflorum* with different area percentage showing the quantitative variation. Except *J. flexile* peak with R_t 3.7 is common in all other species. A peak at R_t 2.2 was observed for *J. auriculatum* & *J. flexile* with area percentage 30.825 % and 73.040 % respectively indicating the higher quantity of that particular component. The species *J grandiflorum* & *J. sambac* showed a compound with R_t 5.03 having area % more than 60. This compound is also present in *J. flexile*, but in small quantity. *J. grandiflorum* and *J. sambac* also showed peaks at R_t 7.1 and 10.0.

J. angustifolium and *J. auriculatum*, showed two peaks (R_t 5.1 & 6.1) as major compounds. Two common peaks with R_t 6.5 & 7.3 were found in both *J. angustifolium* and *J. flexile*. A compound with R_t 6.9 was observed in *J. angustifolium* & *J. sambac* only as traces. A compound with R_t 7.584 was found in *J. auriculatum*, *J. flexile* and *J. sambac.*

The present studies are of highly valuable as it is developed a standard method for the comparative phytochemical analysis such as Thin layer Chromatography, High pressure Liquid Chromatography for the identification of major Phytoconstituents present in different extracts of the selected species. The evaluation of radical scavenging activity with respect to the phenolic compounds present in each species is a major outcome of this present study.

REFERENCES

1. Anonymous. The Wealth of India A Dictionary of Indian Raw Materials and Industrial Products. Publication & Information Directorate, New Delhi: CSIR, 2004: 284-88
2. Bahuguna Y., Juyal V., Rawat M.S.M., Jalalpure S.,‘‘ Diuretic activity of flowers of *Jasminum auriculatum* Vahl,” Journal of Pharmacy Research, 2009; 2 (2): 215-216
3. Ben-Erik Van Wyk & Michael Wink, (2009). *Medicinal Plants of the World*, Briza publication: 22.
4. Chopra R.N., Chopra I.C., Handa K.L., Kapur L.D., Indigenous Drugs of India. Calcutta: U N Dhur & Sons Pvt Ltd, 1958: 512.
5. Cooke T. The Flora of Presidency of Bombay. Vol. II. Calcutta: Botanical Survey of India, 1967:176
6. Deni Bown., The Royal Horticultural Society: Encyclopedia of Herbs and Their Uses,Dorling Kindersly Ltd, London, 1995: 298
7. Deshpande SM; Upadyaya R.R.,‘‘ Chemical studies of *Jasminum auriculatum* (VAHL) leaves,” Curr Sci., 1967; 36: 233
8. Ghosh MN, Fundamentals of experimental pharmacology. Scientific Book Agency, Calcutta, Edition IInd, 1984: 156-157
9. http://www.ehow.com/about_6325794_jasmine-plants-trees.html.
10. Joshi M C., Raju A., Arulanandham A., Saraswathy G.R.,‘‘ Hepatoprotcetive activity of *Jasminum angustifolium* linn. Against ccl4 induced hepatic injury in rat,” Pharmacologyonline, 2008; 3: 197-205
11. Kukic, J., Popovic, V., Petrovic, S., Mucaji, P., Ciric, A., Stojkovic, D., & Sokovic M. (2008). Antioxidant and antimicrobial activity of Cynara cardunculus extracts. Food Chemistry, 107, 861-868
12. Nadkarni A.K. K. M. Nadkarni’s Indian Materia Medica. Vol. I. Bombay: Popular Prakashan Pvt Ltd, 1976: 701
13. Singleton VL and Rossi Jr JA (1965) Colorimetry of total phenolics with phosphomolybdic phosphotungstic acid reagents, *American Journal of Enology and Viticulture*, 16, 144-158

14. Tanahashi T., Nagakura N., Inoue K., Inouye H.,‘‘ Sambacosides a, e and f, novel tetrameric iridoid glucosides from *jasminum sambac*,” Tetrahedron Letters, 1988; 29(15):1793-1796
15. Trivedi, P.C., (2007). *Indian Folk Medicine*, Pointer publishers: 69
16. Zhang Y.J., Liu Y.Q., PU X.Y., Yang C.R.,‘‘ Iridoidal glycosides from *Jasminum sambac*,” Phytochemistry, 1995; 38(4): 899- 903
17. Zhang Z.F., Bian B.L., Yang J., Tian X.F.,‘‘ Studies on chemical constitutents in roots of Jasminum sambac,” Zhongguo Zhong Yao Za Zhi, 2004; 29(3):237-9