

GC- MS analysis of bioactive components of Chloroform and Methanolic leaf extracts of *Justicia glauca*.

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ABSTRACT

The phytochemical constituents of Chloroform and methanol extracts of *Justicia glauca* leaf were analyzed using gas chromatography-mass spectrometry method. The chloroform and methanol leaf extracts of *Justicia glauca* were prepared by soxhlet extraction method and concentrated by using rotary evaporator. The concentrated chloroform and methanol extracts were subjected to phytochemical analysis using GC-MS. GC-MS analysis showed the presence of five and six phytochemicals respectively. The phytoconstituents of chloroform extracts are Hexadecanoic acid methyl ester, Nonadecanoic acid 10- methyl, methyl ester, Stearic acid methyl ester, Methyl Oleate, Z-8,10-dodecadienal. The phytochemicals present in methanol extracts are Hexadecanoic acid methyl ester, 1-H-indene-2-butyl-3-hexyl, Octadecanoic acid methyl ester, E,E,Z-1-3-12-nonadecatriene-5,14 diol, 8H-Azecino[5,4-b] indol-8-one,5-ethylidene- 1,2,3,4,5,6,7,9-octahydro-6-2-hydroxy ethyl)-3- methyl,[S-(E)] and 9-Octadecanoic acid(Z) methyl ester. The result of the GC-MS analysis showed that both the extracts of chloroform and methanol extract of *Justicia glauca* contains many pharmacologically important bioactive compounds. Traditionally, *Justicia* species are used in the treatment of inflammation, gastrointestinal diseases, respiratory tract infection, rheumatism and arthritis. There is need for isolation of these phytochemicals for the therapeutic interest against different diseases.

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Introduction:

Justicia glauca belongs to *Acanthaceae* family. The most important of plant bioactive components are alkaloids, tannins, flavonoids, glycosides, saponins and phenolic compounds. GC-MS studies have been increasingly applied for the analysis of medicinal plants as this technique has proved to be a valuable method for the analysis of non-polar components and volatile essential oils, fatty acids, lipids and alkaloids. Traditionally, *Justicia* species are used in the treatment of inflammation, gastrointestinal diseases, respiratory tract infection, rheumatism and arthritis. Because of their effect on the central nervous system, they are utilized as hallucinogens, sedatives and treatments for epilepsy, depressors, somniferous agents and mental disorders. Some species of *Justicia* are used in the

treatment of diabetes, HIV and cancer. The root, flower, fruit and leaf of *J. adhatoda* have been used in the treatment of bronchitis, cold, whooping cough, asthma, helminthic, diarrhoea, dysentery, and glandular tumor, expectorant, tuberculosis, abortifacient, microbial, tussive and cancer. Decoction of *J. anselliana* leaves and roots are used in the treatment of heart diseases and testicles inflammation. The constituents of *J. spicigera* are potassium, calcium acetate, oxalate, sulfate, sodium chloride, simple carbohydrates, mucilages, pectins, glycosides, pigments, resin and essential oils. Flavonoids have been isolated from the leaves of *J. spicigera*. The total phenols, flavonoids and antioxidant activity of *J. Spicigera* was reported by Sepúlveda-Jiménez and co-workers in 2009. Euler and Adam, in

1982 suggested the presence of triterpenoids of kaempferol and kaempferitrin by characterization based on NMR, GC-MS and UV analysis of *J. spicigera* extracts. The presence of β - sitosterol, 3-O - glucoside of β - sitosterol and cryptoxanthin in hexane-isopropyl ether-methanol extracts of *J. spicigera* was identified by Domínguez et. Similarly six compounds were identified in ethanolic extract of *J. carnea* which have properties like vascular relaxant, acidifiers, aiding in cell division and chelation of heavy metal. Presently, the interest in studying and understanding the constituents of plants with healing properties has increased. The interest is focused in obtaining biologically active molecules and using them for curing various diseases. In continuation with the ongoing research on phytochemicals of *Justicia* species, we hereby present GC-MS analysis of ethanol extract of *Justicia Glauca*.

Methodology:

Plant Material:

Fresh leaves of *Justicia glauca* were collected from the regional areas of Srikakulam, Andhra Pradesh. The plant was identified at the Taxonomy section of Department of Botany, Andhra University, Visakhapatnam, Andhra Pradesh, India.

Preparation of Plant Extract:

Justicia glauca was dried in a shady place for 10 days and coarsely powdered. Extraction was obtained using soxhlet method. Approximately 45gm of the powdered plant material was introduced into the extraction chamber of the soxhlet extractor, using chloroform and methanol as solvents (based on the polarity) and the extraction was done. At the end of the extraction, the extracts were concentrated using rotary

evaporator and concentrated extract was carefully stored for further analysis. Dried extract was used for GCMS analysis.

GC-MS analysis of *Justicia Glauca*:

The phytochemical investigation of methanol extract of *J. adhatoda* leaf was performed on a GC-MS equipment (Thermo Scientific Co.) Thermo GC-TRACE ultra ver.:2.2, Thermo TSQ QUANTUM XLS. Experimental conditions of GC-MS system were as follows: DB 5-MS capillary standard non-polar column, dimension: 30Mts, ID: 0.25 mm, Film thickness: 0.25 μ m. Flow rate of mobile phase (carrier gas: He) was set at 1.0 ml/min. In the gas chromatography part, temperature programme (oven temperature) was 40 °C raised to 290 °C at 5 °C/min and injection volume was 1.0 μ l. A scan interval of 0.5 seconds with scan range of 40-600 m/z. Total GC running time was 35min. The identification of compounds was based on comparison of their mass spectra with those of WILEY and NIST Libraries.

Identification of Phytocomponents in *Justicia glauca*

The retention indices, peak area percentage and mass spectra fragmentation pattern of GC-MS chromatogram of ethanol extract of *Justicia glauca* was compared with the database of National Institute of Standards and Technology (NIST), NIST08.LIB, WILEY8.LIB and with published literature. The name, molecular weight, formula, structure and bioactivities of the compounds were ascertained.

Results and Discussions:

The bioactive components of chloroform and methanol extract of *Justicia Glauca* is represented in both tabular form and also in Gas Chromatogram.

Table 1: Identification of chemical constituents of Chloroform leaf extracts of *J. glauca* by GC-MS

S. No	Retention Time	Peak area	Molecular formula	Molecular weight	% Peak area
1.	17.1	Hexadecanoic acid methyl ester	C ₁₇ H ₃₄ O ₂	270.45	62.4
2.	20.98	Nonadecanoic acid 10-methyl, methyl ester	C ₂₀ H ₄₀ O ₂	312.53	7
3.	19.07	Stearic acid, methyl ester	C ₁₉ H ₃₈ O ₂	298.51	64.4
4.	18.83	Methyl oleate	C ₁₉ H ₃₆ O ₂	296.49	51
5.	16.18	Z-8,10-dodecadienal	(-)	(-)	2.9

Table 2: Identification of chemical constituents of Methanol leaf extracts of *J. glauca* by GC-MS

S. No	Retention Time	Peak area	Molecular formula	Molecular weight	% Peak area
1.	17.1	Hexadecanoic acid methyl ester	C ₁₇ H ₃₄ O ₂	270.45	64.1
2.	18.18	1H-indene-2-butyl_3_hexyl	C ₁₉ H ₂₈	256.42	2.5
3.	19.07	Octadecanoic acid, methyl ester	C ₁₉ H ₃₈ O ₂	298.5	54.1
4.	19.77	E,E,Z-1-3-12-nonadecatriene- 5,14 diol 8H-Azecino[5,4-b] indol-8-one,	C ₁₉ H ₃₄ O ₂	297.47	4.9
5.	18.6	5-ethylidene- 1,2,3,4,5,6,7,9-octahydro -6-2-hydroxy ethyl)-3- methyl,[S-(E)]	(-)	326	7.7
6.	18.82	9-Octadecanoic acid(Z) methyl ester	CH ₃₆ O ₂	296.4	53.8

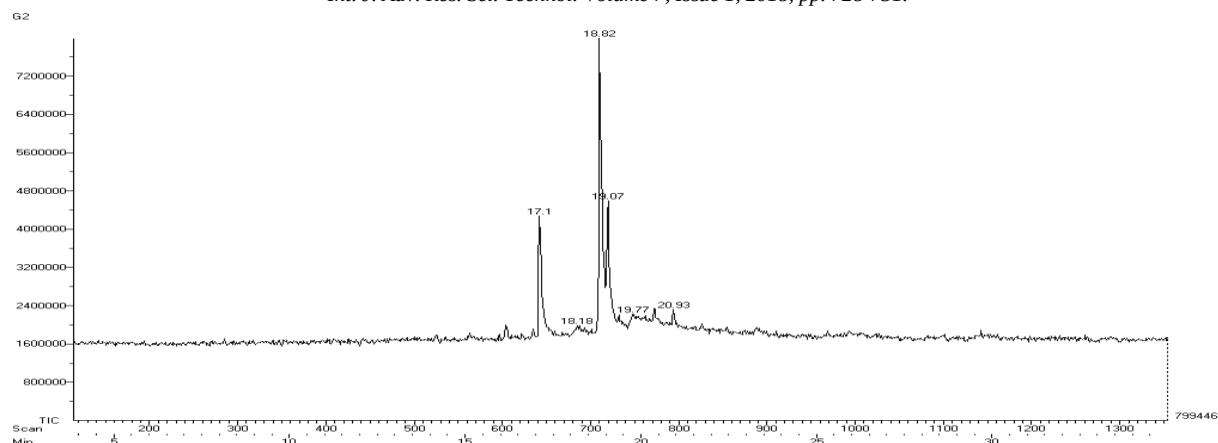


Fig. 1: GC-MS Analysis of Chloroform leaf extract of *Justicia glauca*.

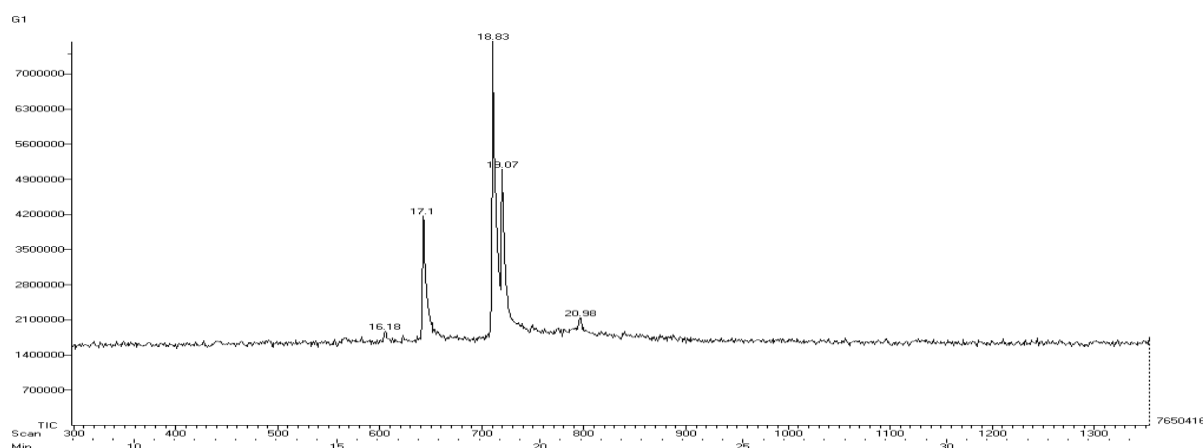


Fig. 2: GC-MS Analysis of Methanol leaf extract of *Justicia glauca*

In Table 1 it is shown that the chloroform leaf extract of *Justicia glauca* contains five chemical constituents and Table 2 shows that methanol leaf extract contains six chemical constituents. In both the extracts Hexadecanoic acid methyl ester is common constituent which functions as antioxidant, pesticide, flavor agent, 5-Alpha-Reductase inhibitor, anti-fibrinolytic, hemolytic, lubricant, nematocide and atialopepic. This is also one of constituents of methanol stem and leaf extract of *J. wynaadensis* and methanolic leaf extract of *J. adathoda*. E,E,Z-1-3-12-nonadecatriene-5,14 diol has antibacterial property. 8H-Azecino[5,4-b] indol-8-one, 5-ethsylidene-1,2,3,4,5,6,7,9-octahydro-6-2-hydroxy ethyl)-3- methyl,[S-(E)] is anew compound which is also found in methanol seed extract of *Lepidium Sativum*, 9-Octadecanoic acid(Z) metyhl ester is a flavoring agent and adsorbent it is also found in petroleum ether extract of the stem of *J. adathoda*. Octadecanoic acid methyl ester is found in cloves. It acts as antifoaming agent and fermentation nutrient. Methyl oleate is used as antifoaming and food additive.

Conclusion:

In the present study, some of the constituents have antioxidant, antibacterial and activities. Activities for other compounds need further pharmacological investigation in order to develop new drugs for treating

specific diseases. Thus GC-MS analysis of *Justicia glauca* confers that the plant is medicinally potential and paves a way for further research.

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