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PHYTOCHEMICAL SCREENING AND GC-MS ANALYSIS OF ROOT EXTRACT FROM *ASPARAGUS RACEMOSUS L.*

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ABSTRACT: The present chemical analysis deals with *Asparagus racemosus* root for phytochemical screening and GC-MS analysis. Phytochemical constituent of the root extract was carried out according to the standard method. GC-MS analysis was performed on the methanolic extract of *A. racemosus* to find out the chemical constituents. The phytochemical screening revealed the presence of flavonoids, alkaloids, steroids, terpenoids, glycosides, tannins, terpenoids, saponins. GC-MS results revealed the presence of 6 different phytochemical constituents like, 2-Propanone, 1,3-dihydroxy 1), 2-Fruancarboxy aldehyde, 5-(hydroxymethyl) 2), Hexadecanoic acid 3), n-Hexadecanoic acid 4), Ethanol,2(Octyloxy)- 5), 1,9-Nonanediol 6). The presence of various biological active compounds confirms the application of *A. racemosus* for various diseases by conventional practitioners. The obtained phytochemical constituent results demonstrate that the methanol root extract of *Asparagus racemosus* could be used corn foot, anticancer, diabetic healer. However, isolation of individual phytochemical constituents might be useful to formulate a novel drug.

INTRODUCTION: Herbal medicines have been used for ancient time to present day for human diseases¹. Medicinal plants are equipped from a diversity of plant materials as leaves, stems, roots, barks, inflorescence etc². Medicinal plants contain biologically active principles with specific therapeutic effects. *Asparagus racemosus L.* that belongs to Asparagae is a small perennial climber found all over different parts of Tamil Nadu, India. This climber has been used as a source of medicine by human from Ancient time. *A. racemosus* is an important herbal climber and it is widely used as Rasayana in Ayurvedic systems of medicines³.

According to ancient classical Ayurvedic literature claimed several therapeutic attributes for the root of *A. racemosus* and has been specially recommended in cases of galactogogue⁴. The traditional knowledge to scientific evidence was found to be lacking due to the traditional information that was kept secret by the village vaidyas⁵.

Modern pharmaceutics also have been benefited from herbal plants. The specific healing effect of herbal plants contains several phytochemical constituents^{6,7}. The major bioactive components in the roots are alkaloids, a diverse class of natural products. A wide variety of secondary metabolites inclusive of alkaloids, steroids and flavonoids are responsible for therapeutic properties of plants⁸. The therapeutic achievements of the herbal plants exclusive to particular plant species or groups are consistent with the concept that the combination of secondary products in a particular plant is

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taxonomically distinct⁹. Medicinal plants provide health care and disease free life. Biologically active principles contained in it are exploited in conventional medicine for various ailments¹⁰. The spectrometry and chromatographic screening method could provide the needed preliminary observations to select crude plant extracts with potentially useful properties for further chemical and pharmacological investigation. The determination of phytochemical constituents is mostly performed by the moderately expensive and often laborious techniques such as gas (GC) and liquid chromatography (LC) combined with specific detection schemes¹¹. In the past few years, GC-MS has become strongly established as a key technological metabolic profiling in both plant and non-plant species^{7, 12}. The crude extracts and medicines manufactured of the principles of biological active compounds even by pharmaceutical companies could be lead to large scale exposure of humans to natural products.

In regulate to encourage the use of medicinal plants, it should be thoroughly investigated with their composition, activity and thus validate their use¹³. To the best of our knowledge, there is no scientific evidence to justify the traditional use of *A. racemosus* root in the ailment of corn foot. The present investigation, of this work was to study the natural bioactive products and their phytochemical constituents, corn foot activities in organic solvent extracts prepared from the *Asparagus racemosus*.

MATERIALS AND METHODS:

Collection of plant materials

The roots of *Asparagus racemosus* were collected from traditional healers in Elusempon, Villupuram District, Tamil Nadu, India during January - 2012.

Chemicals

All chemicals were purchased from SD fine chemical company Mumbai and all chemicals were of analytical grade.

Preparation of root extract

In the laboratory, fresh root were cut into small piece and dried then powered with a mechanical grinder and stored in a jib lock cover. The dried powder (100g) was refluxed with methanol for seven days. Removal of whole dry plants was done by filtration through whatman No.1 filter paper.

The extracts were placed in glass petridishes. The total filtrate was concentrated to dryness, in hot air oven at 32°C to render methanol solvent extract. The dried crude extract (blackish-green yield 5.5% w/w) was found which was then re-dissolved in methanol to obtain a solution containing 2.0mg/ml of extracts to be used for further assays.

Qualitative phytochemical group test

Asparagus racemosus root extract was subjected to qualitative screening for the detection of phytochemical groups by standard method¹⁴.

Gas chromatography-Mass spectrometry (GC-MS) analysis

The analysis of the root powder were performed using a Hewlett Packard 5890 II GC, equipped with a HP-5 MS capillary column (30m.25, film thickness 0.251m) and a HP5972 mass selective detector. For GC-MS detection an electron ionization system with ionization energy of 70 eV was used. Helium was the carrier gas, at a flow rate of 1m/min. Injector and MS transfer line temperature were set at 220 and 290° C respectively. The diluted samples (1/100 in acetone, v/v) of 1ul were injected manually and in the splitless mode. The components were identified based on the comparison of their relative retention time and mass spectra with those of standards, NBS75K library data of the GC-MS system and literature data. The results were also confirmed by the comparison of the compounds elution order with their relative retention indices on non-polar phases reported in the literature.

RESULTS AND DISCUSSION:

Qualitative phytochemical screening of *A. racemosus* root extract Methanol extract of *A. racemosus* root showed the presence of alkaloids, steroids, flavanoids, glycosides, saponins, terpenoids, (Table 1).

GC-MS Analysis of *A. racemosus* root extract The results shows to GC-MS analysis lead to the identification of number of compounds from GC fractions of the methanolic extract of *A. racemosus*. They were identified through mass spectroscopy attached with GC. GC-MS analysis of methanolic extract of *A. racemosus* was put into a (Table 2). The results revealed the presence of 6 different phytocompounds viz., 2-Propanone, 1, 3-dihydroxy

– (10.97%), 2-Fruancarboxy aldehyde, 5-(hydroxymethyl) - (76.60%), Hexadecanoic acid- (2.17%), n-Hexadecanoic acid- (4.49%), Ethanol, 2 (Octyloxy) - (3.91%), 1, 9-Nonanediol- (1.86%). The GC-MS spectrum confirmed six compounds from *A. racemosus* root were identified by the presence of 6 components with the retention time 5.301, 8.719, 15.069, 17.129, 20.139, and 21.609 respectively. The details of the identified phytoconstituents and its therapeutic activity were given in the (Table 3). The retention time, compound name, molecular formula, molecular structure, extracted ions of the compounds were

examined. GC-MS analysis of methanolic extract of *A. racemosus* was tabulated in (Table 4). Medicinal plants in all facts of life have served a essential starting material for drug design and development¹⁵. Antifungal and antibacterial substances found to saponine, flavonoids, glycosides and alkaloids etc are found to be distributed in plants, yet these compounds were not well established due to the lack of knowledge and techniques¹⁶. From this phytochemical constituent, saponins have been reported to exhibit haemolytic and foaming activity¹⁷.

TABLE.1. PHYTOCHEMICAL SCREENING OF METHANOL ROOT EXTRACTS OF *A. RACEMOSUS*.

	Phytoconstituents	Name of the test	Observed changes	Result
1.	Flavonoids	Ammonium test Dragendorff's test	Yellow color Red precipitate	+ +
2.	Alkaloids	Wagner's test Hagers test	Reddish-brown precipitate Yellow precipitate	+ +
3.	Steroids	General test	Reddish brown	+
4.	Terpenoids	General test	Grey color	+
5.	Glycosides	Fehling's test	More dense red	+
6.	Tannins	Lead acetate test Ferric chloride test	Cream gelatinous precipitate Greenish to black color	+ +
7.	Saponins	Frothing test	Stable froth	+

TABLE 2. IDENTIFIED PHYTOCHEMICALS FROM THE METHANOL ROOT EXTRACT OF *A. RACEMOSUS*

S.No	Retention time	Name of the compounds	Molecular formula	Molecular weight	Area(%)
1	5.301	2-Propanone, 1,3-dihydroxy	C ₃ H ₆ O ₃	90.00	10.97
2	8.719	2-Fruancarboxy aldehyde, 5-(hydroxymethyl)	C ₆ H ₆ O ₃	126.00	76.60
3	15.069	Hexadecanoic acid	C ₁₉ H ₃₈ O ₄	330.50	2.17
4	17.129	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	4.49
5	20.139	Ethanol,2(Octyloxy)-	C ₁₀ H ₂₂ O ₂	174.00	3.91
6	21.609	1,9-Nonanediol	C ₉ H ₂₀ O ₂	160.00	1.86

TABLE 3.THERAPEUTIC ACTIVITY OF THE PHYTOCOMPOUNDS CONSTITUENTS IDENTIFIED FROM THE METHANOL ROOT EXTRACT OF *A. RACEMOSUS* ROOT BY GC-MS.

S.No	Name of the compound	Therapeutic Activity
1	2-Propanone, 1,3-dihydroxy	Do not induce vomiting, artificial respiration, antioxidant.
2	2-Fruancarboxy aldehyde, 5-(hydroxymethyl)	Antioxidant, antiacne Antimicrobial and Preservative, Antibacterial, Antitumor, Cancer-Preventive,
3	Hexadecanoic acid	Lubricant,antiandrogenic,antioxidant, 5-alpha-reductase inhibitor
4	n-Hexadecanoic acid	Antioxidant, hypocholesterolemic nematocide, pesticide, anti-androgenic flavor, hemolytic and 5-Alpha reductase inhibitor, Hemolytic
5	Ethanol,2(Octyloxy)-	Antioxidant, antibacterial
6	1,9-Nonanediol	Not reported
7	2-Furaldehyde	Antimicrobial and Preservative, Antibacterial, Antioxidant, Antitumor, Cancer-Preventive,
8	2-Furaldehyde, 5-hydroxyle	Antibacterial, Antioxidant, Antitumor,
9	Hydrazine, 1,1-dimethyl	Anti-androgenic flavor, haemolytic, Antioxidant.
10	Ethanimidic acid	hypocholesterolemic nematocide, pesticide, anti-androgenic flavor, hemolytic and 5-Alpha reductase inhibitor

** Source:Dr. Duke's phytochemical and ethano botanical databases [Online database].

TABLE 4. THE EXTRACTED IONS WITH THEIR RELATIVE RETENTION TIMES DURING THE GC-MS ANALYSIS

RT	Compound Name	Formula	m/z
5.301	2-Propanone, 1,3-dihydroxy		97
8.719	2-Fruancarboxy aldehyde, (hydroxymethyl)		126
15.069	Hexadecanoic acid		129
17.129	n-Hexadecanoic acid		256
20.139	Ethanol,2(Octyloxy)-		129
21.609	1,9-Nonanediol		129
8.717	2-Furaldehyde		126
8.716	2-Furaldehyde, 5-hydroxyle		126
7.121	Hydrazine, 1,1-dimethyl		125
7.892	Ethanimidic acid		124
15.067	Tetradecanoic acid		228

Generally, hereby plants are essential plant material having a wide range of local healing applications, the leaves, roots, barks and seeds are found to be antipyretic, laxative, analgesic, antioxidant, antifungal, antibacterial and non inflammatory¹⁸. Active phytochemical investigation revealed that the extracts contained a variety of phytoconstituents. Saponins, tannins, alkaloids and, flavonoids are present in the root extracts. The

bioactive compounds including thiocyanate, nitrate chloride and sulphates, beside other water soluble components which are naturally occurring in most plant materials, are known to be bacterial, pesticidal or fungicidal in nature thus conferring the antimicrobial property to plants^{19, 20}. From this phytochemical constituent, saponins have been reported to exhibit hemolytic and foaming activity²¹. To the best of my knowledge,

compounds 1-6 were isolated from *Asparagus racemosus*.

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