

GC-MS analysis of phytochemicals in the methanolic extract of *Oldenlandia umbellata*

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ABSTRACT

The aim of this study was to carry out Gas chromatography and Mass spectroscopy analysis of phytochemicals in the methanolic extract of *Oldenlandia umbellata*. The GC-MS analysis of *O. umbellata* powder extract with in methanol is performed using GC-MS equipment Thermo GC-TRACE ultra ver: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system are as follows: DB 35 - MS capillary standard non - polar column, dimension: 30 Mts, ID: 0.25 mm, FILM: 0.25 µm is used and flow rate of mobile phase (carrier gas: He) is set at 1.0 ml/min. GC-MS chromatogram of methanolic extract of *O. umbellata* shows 22 peaks indicating the presence of twenty two compounds. GC-MS analysis reveal that the presence of 22 different phytochemical compounds namely Heptacosyl pentafluoropropionate - (17.34%), Cholestane 3,6,7-triol, (3,5,6,7-tetrahydro) - (2.54%), Methane, oxybis [dichloro - (1.09%), Dibutyl-phthalate-(0.16%), Phytol-(0.11%), Nonacosane-(0.11%). GC-MS analysis shows the existence of various compounds with different chemical structure. Thus, this type of GC-MS analysis is the first step towards understanding the nature of active principles in this medicinal plant and this type of study will be helpful for further detailed study.

Keywords: GC-MS analysis, *Oldenlandia umbellata*, Methanolic extract.

1. INTRODUCTION

The medicinal plants are of great interest to human health. Plant based medicines have been a part of traditional healthcare in most parts of the world for thousands of years [1, 2]. Many medicinal plants are used daily in Ayurvedic practices. In India more than 7,000 medicinal plant species are known. The medicinal plants find application in pharmaceutical, cosmetic, agricultural and food industry. Plants contain numerous biologically active compounds, many of these have been shown to exhibit antimicrobial properties and therefore they were in use as antimicrobial drugs in traditional medicines. According to a report of World Health Organization, more than 80% of world's populations depend on traditional medicine for their primary health care needs [3]. Fairly comprehensive information of the curative properties of some of the herbs has been recorded in "charak samhita" and "sushruta samhita" [4].

Knowledge of the phytochemical is desirable not only for the discovery of healthcare products but also in disclosing new sources of

economic materials like alkaloids, tannins, oils, gums etc., [5]. The systematic screening of plant extracts or plant derived substances still remains an interesting strategy to find new lead compounds in many plant species. So the determination of phytoconstituents is largely performed by the relatively expensive and often laborious techniques such as gas (GC) and liquid chromatography (LC) combined with specific detection schemes [6]. In the last few years, GC-MS has become firmly established as a key technological metabolic profiling in both plant and non plant species [7-9]. The genus *Oldenlandia* (family Rubiaceae) consists of different species, many of which are used in traditional medicine [10]. *Oldenlandia umbellata* is commonly known as "Indian madder", known to yield a color-fast red dye from its roots, and has been used in diverse applications since ancient times. The root bark, preferably of a two-years-old plant, when used with a mordant will confer red color to calico, wool, and silk fabrics [11]. It is a low growing plant native to India and commonly found in parts of

India (Coromandel coast), Burma, Sri Lanka, Cambodia and Indonesia. The plant is well-known in Siddha Medicine for its styptic property. It is also a drug that can be administered for bronchial asthma, as a decoction of the entire plant, a decoction made from its root and liquorice in the ratio 10:4 or the powdered root is given either with water or honey. Both leaves and roots are also deemed good expectorants, and used for treatment of asthma, bronchitis, and bronchial catarrh [12]. In folklore medicine this plant is widely used in the treatment of various ailments. The decoction of the plant is widely used as an expectorant and febrifuge. It is also used in treatment of cancer, asthma and tuberculosis [13-14]. These varied uses have increased utilization and exploitation of *O. umbellata* for medicinal and dye extraction purposes [15]. As a result, natural stands of *O. umbellata* are fast disappearing and are threatened with extinction due to indiscriminate collection. The plant grows wild in forests, among other areas, and there is no propagation system available to replenish these stands. Therefore, an effort has been undertaken to develop a reliable protocol for mass multiplication of *O. umbellata* in present investigation. Literature survey of this medicinal revealed that no extensive phytochemical and pharmacological investigations had been carried out.

2. MATERIAL AND METHODS

2.1. Collection of the plant material

The plant *Oldenlandia umbellata* was collected from Thothukudi district (Tamil Nadu), India, during the months of October and November 2011 and all the primary work done (washing, drying...etc.). The plant materials were identified and authenticated by Dr. V. Chelladurai, Retired Research officer-botany, Central Council for Research in Ayurveda and Siddha (C.C.R.A.S). Govt. of India, Tirunelveli. The collected plant material was free from disease and also free from contamination of other plants.

2.2. Preparation of plant extract

100g of *O. umbellata* air dried and coarsely powdered entire plant material was extracted with 500ml methanolic solvent by using Soxhlet extractor. After extraction the sample were kept in dark for 72 hrs with intermittent shaking. The solvent was decanted and distilled off in Rotovac apparatus. The methanol extract was completely dried from solvent under reduced pressure using high vacuum conditions. The collected extract was then taken up for further investigations.

2.3. GC-MS analysis

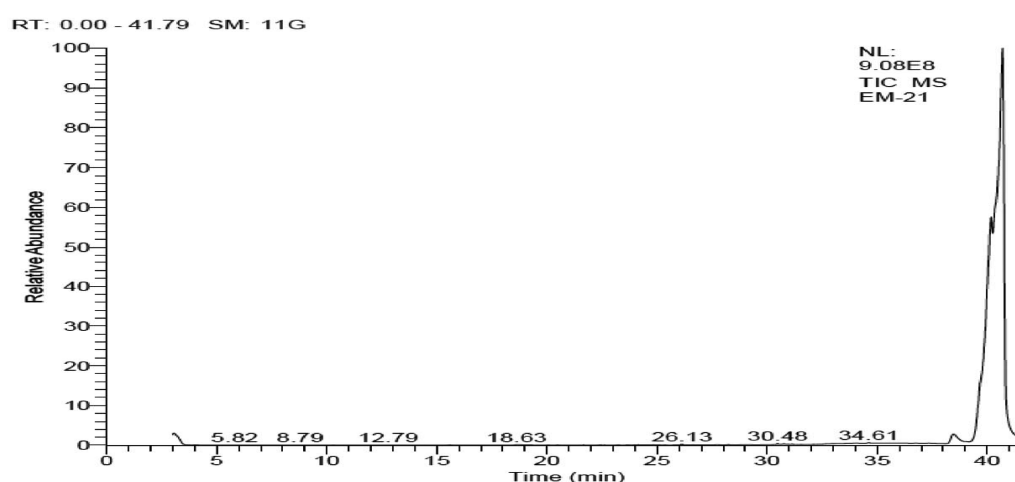
The GC-MS analysis of *O. umbellata* powder extract with in methanol was performed using GC-MS equipment Thermo GC-TRACE ultra ver: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system were as follows: DB 35 - MS capillary standard non-polar column, dimension: 30 Mts, ID: 0.25 mm, FILM: 0.25 μ m was used and 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 oven temp 50 c raised to 260 c at 10 c/min and injection volume was 1 micro litre. Samples which dissolved in methanol were run fully at a range of 50-650 m/z and the results were compared by using Wiley Spectral library search programme. The Mass Spectra detected in 38.79 Min.

3. RESULTS AND DISCUSSION

GC-MS chromatogram of methanolic extract of *O. umbellata* showed 22 peaks indicating the presence of twenty two compounds. The chemical compounds identified in the methanol extract of the *O. umbellata* presented in Table 1. GC-MS analysis revealed that the presence of 22 different phytochemical compounds namely Methane,oxybis[dichloro - (1.09%), Cyclopentane carboxylic acid, 3-(3-fluorophenylcarbonyl)-1,2,2-trimethyl - (0.04%), 1-Nitro-2-acetamido-1,2-dideoxy-d-glucitol - (0.06%), 2-(2-Methylpropenyl) - cyclohexanone - (0.03%), 6-Methylcyclodec-5-enol - (0.03%), Metanephrene - (0.03%), Phenol, 3-methyl-4-(methylthio) - (0.04%), 9,10-Dimethyltricyclo[4.2.1.1(2,5)]decane-9,10-diol - (0.05%), Dibutyl phthalate - (0.16%), Phytol - (0.11%), Oleic acid, eicosyl ester - (0.04%), 7,8-Epoxylanostan-11-ol, 3-acetoxy - (0.03%), Dasycarpidan-1-methanol, acetate (ester) - (0.03%), Trilinolein - (0.03%), 4,13,20-Tri-O-methylphorbol 12-acetate - (0.03%), Nonacosane - (0.11%), Cholestane-3,5-dichloro-6-nitro-, 3 α ,5 α ,6 α - (0.03%), 2,2-Bis(4-propionyloxyphenyl)propane - (0.04%), 2-Ethylamino-3-methyl(phenyl)amino-1,4-naphthoquinone - (0.04%), Etocrylene - (0.08%), Cholestane-3,6,7-triol, (3 α ,5 α ,6 α ,7 α) - (2.54%), Heptacosyl pentafluoropropionate - (17.34%). The GC-MS spectrum confirmed the presence of 22 components with the Retention Time 3.10, 15.58, 18.62, 22.43, 24.13, 24.84, 27.72, 28.24, 30.48, 30.76, 30.96, 31.56, 32.11, 32.92, 34.01, 34.61, 35.10, 36.30, 36.73, 37.33, 38.45 and 40.14 respectively (Figure 1). Carbohydrates like allose and sucrose are considered amount is present. The GC-MS analysis revealed that the methanolic extract is mainly composed of oxygenated hydrocarbons and predominantly phenolic hydrocarbons. These phytochemicals are

Table 1: Compounds present in the methanolic extract of *Oldenlandia umbellata* using GC-MS analysis.

RT	Name of the Compound	MF	MW	Peak area %
3.10	Methane,oxybis[dichloro-	C2H2Cl4O	182	1.09
15.58	Cyclopentanecarboxylicacid, 3-(3-fluorophenylcarbamoyl)-1,2,2-trimethyl	C16H20FN03	293	0.04
18.62	1-Nitro-2-acetamido-1,2-dideoxy-d-glucitol	C8H16N2O7	252	0.06
22.43	2-(2-Methyl-propenyl)-cyclohexanone	C10H16O	152	0.03
24.13	6-Methyl-cyclodec-5-enol	C11H20O	168	0.03
24.84	Metanephrine	C10H15NO3	197	0.03
27.72	Phenol, 3-methyl-4-(methylthio)	C8H10OS	154	0.04
28.24	9,10Dimethyltricyclo[4.2.1.1(2,5)]decane-9,10-diol	C12H20O2	196	0.05
30.48	Dibutyl phthalate	C16H22O4	278	0.16
30.76	Phytol	C20H40O	296	0.11
30.96	Oleic acid, eicosyl ester	C38H74O2	562	0.04
31.56	7,8-Epoxyloganostan-11-ol, 3-acetoxy	C32H54O4	502	0.03
32.11	Dasycarpidan-1-methanol, acetate (ester)	C20H26N2O2	326	0.03
32.92	Trilinolein	C57H98O6	878	0.03
34.01	4,13,20-Tri-O-methylphorbol 12-acetate	C25H36O7	448	0.03
34.61	Nonacosane	C29H60	408	0.11
35.10	Cholestane-3,5-dichloro-6-nitro-, 3 α ,5 α ,6 α -	C27H45Cl2NO2	485	0.03
36.30	2,2-Bis(4-propionyloxyphenyl)propane	C21H24O4	340	0.04
36.73	2-Ethylamino-3-methyl(phenyl)amino-1,4-naphthoquinone	C19H18N2O2	306	0.04
37.33	Etocrylene	C18H15NO2	277	0.08
38.45	Cholestane-3,6,7-triol, (3 α ,5 α ,6 α ,7 α)	C27H48O3	420	2.54
40.14	Heptacosyl pentafluoropropionate	C29H55F3O2	492	17.34

**Figure - 1: GC-MS chromatogram of methanolic extract of *Oldenlandia umbellata***

responsible for various pharmacological actions like antimicrobial activity. This study is only preliminary study of the occurrence of certain properties of *Oldenlandia umbellata* extract an in-depth study will provide a good concrete base for all the biochemical and phytochemical functions

mentioned above. New scientific strategies for the evaluation of natural products with specific biological activities require the implementation of large screen process.

Oldenlandia umbellata is a potential folklore medicinal plant use for many diseases and

infections. Phytochemical analysis by GC-MS revealed the presence of fatty acid ester, fatty acid amide, terpenoids, phytol and pentafluoropropionate like important constitution. In this study, for the first time, a convenient and sensitive GC-MS method was developed for identification and quantification of twenty two characteristic components in *O. umbellata*. GC-MS analysis of methanolic extract of *Oldenlandia umbellata* was tabulated in (Table 1).

4. CONCLUSION

GC-MS analysis showed the existence of various compounds with different chemical structure.

Thus, this type of GC-MS analysis is the first step towards understanding the nature of active principles in this medicinal plant and this type of study will be helpful for further detailed study.

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