**GC-MS Analysis of Methanol and Ethyl Acetate Extract of fruits of Sphaeranthus indicus**

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**ABSTRACT**

**Introduction:** To identify the various phytoconstituents present in the plant *Sphaeranthus indicus* by using gas chromatography and mass spectrometry. **Methods:** The fruits of *Sphaeranthus indicus* were extracted with different solvents of increasing polarity. The methanol and ethyl acetate extract were subjected to GCMS analysis to detect the phytoconstituents. **Results:** Totally 26 compounds were identified. Among these 13 constituents in methanol extract and 13 constituents in ethyl acetate extract were identified during the GC-MS analysis. Stigmasterol and lupeol which were identified in the plant is considered to have antiarthritic properties. **Keywords:** *Sphaeranthus indicus*, Gas chromatography, Mass spectrometry.

**INTRODUCTION**

Plants are considered as useful source of medicines from the thousands of years. About four million people use the traditional system of medicine or herbal medicines for primary health care. In the present scenario, the understanding of the chemical constituents of plants with medicinal properties play a crucial role in identifying new source of economically viable phytoconstituents. The study also helps in validating the actual significance of the traditional medicinal practices. Hence thorough understanding of these chemical constituents became the key focus area in standardizing the natural drug because many of these phytoconstituents exhibits a complementary and overlapping mechanism of activity. Mass spectrometry, along with chromatographic separation technique such as Gas chromatography (GC/MS) is usually used in case of direct analysis of the components existing in traditional medicinal practices and medicinal plants.

*Sphaeranthus indicus* (L.) commonly known as Gorakhmundi belongs to family Asteraceae. It is a medicinally important plant used as folk medicine. It is an important medicinal plant used for the treatment of gastric disorders, skin diseases, anthelmintic, glandular swelling, nervous depression, analgesic, antibiotics, antifungal, laxative and diuretic properties. The powdered bark along with whey is useful in the treatment of piles. Flowers have alterative, depurative and stimulant characters. Roots and seeds are anthelmintic. Juice of fresh leaves is taken for cough. The plant is also useful in preservation of food grains as it possess insecticidal property. In the present study, we evaluated the phytoconstituents of methanol and ethyl acetate extract of *Sphaeranthus indicus* fruits by gas chromatography and mass spectrometry (GC-MS) to provide the scientific information to develop potential phytomedicine.

**MATERIALS AND METHODS**

**Collection and Authentication of Plant Material**

Fruits of *Sphaeranthus indicus* was collected and certified from organic botanical garden of Kurukshetra University, India, during March. The voucher specimens were identified by a taxonomist, by Dr. B. D. Vashist, Department of Botany, Kurukshetra University, Kurukshetra, with reference no. KUK/BOT/IPS 33. The voucher specimens KUK/BOT/IPS 33 were deposited at the Department of Pharmaceutical Sciences, Kurukshetra University, Haryana, India, for future reference.

**Preparation of fruit Extract**

The plant material was washed and rinsed with tap water and dried in shade. The powdered plant material was extracted with methanol using a Soxhlet extractor and then it was
is successively extracted with ethyl acetate for 72 h and the extract obtained was filtered and concentrated using rota evaporator.

**GC-MS analysis**

GC-MS analysis was carried out on an Agilent system equipped with Mass Spectrometer detector and split/splitless injection system. The GC was fitted with a HP-5MS capillary column (30m X 250m; film thickness: 0.25m). The temperature program was as follows: injector temperature 280°C, initial oven temperature at 50°C, then increased at 25°C/min to 300°C and was hold for 10 mins. Helium was used as carrier gas at 17.69 psi pressure with flow 2.1 ml/min. Samples were dissolved in methanol and 1 μl aliquot were injected automatically. MS spectra of separated components were identified based on WILEY and NIST Libraries for botanical compounds.

GC-MS detection of phytoconstituents in methanol and ethyl acetate extract of *Sphaeranthus indicus* were based on the computer evaluation of mass spectra of samples through comparison of peaks and retention time.

**RESULTS AND DISCUSSION**

GCMS is one of the most precise methods to identify various secondary metabolites present in the plant extract. Totally 26 compounds were identified. Among these 13 constituents in methanol extract (Table 1) and 13 constituents in ethylacetate extract (Table 2) were identified during the GC-MS analysis. The chromatograph of methanol extract and ethyl acetate extract showed different peaks (Figure 1 and Figure 2). The constituents identified in the methanol extract was 1,2-Benzenediol (9.54%), 22,23-dihydro Stigmasterol (1.76 %), Stigmasterol (0.32%). The constituents identified in the ethylacetate extract was 2,4-Decadienal (9.54%), 3-Octadecene(4.07%), lupeol (0.57%). Many other compounds were also identified in both the extracts.

**CONCLUSION**

The present study of the analysis of two different extracts methanol and ethyl acetate has suggested the presence of 26 of phytocompounds. Among these some compounds are responsible for antiarthritic activity (Stigmasterol and lupeol). Thus the medicinal plant *Sphaeranthus indicus* is found to possess significant phytoconstituents responsible for antiarthritic activity. The presence of such variety of phytochemicals may justify the use of the plant in the traditional system of medicine for treating rheumatoid arthritis.
Table 1: Phytoconstituents of methanol extract of *Sphaeranthus indicus*

| No | RT (min) | Name of the Compounds | Molecular Formula | Molecular Weight | Height% |
|----|----------|-----------------------|------------------|-----------------|--------|
| 1  | 10.583   | 4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl | C_{4}H_{6}O_{4} | 144 | 0.30 |
| 2  | 10.618   | 1,2-Benzenediol       | C_{6}H_{4}O_{2} | 110 | 9.54 |
| 3  | 10.725   | Phenol,2,6-dimethoxy  | C_{6}H_{10}O_{3} | 154 | 0.47 |
| 4  | 10.837   | Methylparaben         | C_{6}H_{12}O_{2} | 152 | 0.80 |
| 5  | 11.077   | Phenol,2,4-bis(1,1-dimethylethyl) | C_{8}H_{24}O_{12} | 206 | 0.95 |
| 6  | 11.280   | Caffeine              | C_{10}H_{34}N_{2}O_{2} | 194 | 4.07 |
| 7  | 11.964   | Hexadecanoic acid, methyl ester | C_{16}H_{34}O_{2} | 270 | 0.73 |
| 8  | 12.064   | n-Hexadecanoic acid  | C_{16}H_{32}O_{2} | 256 | 0.38 |
| 9  | 12.123   | Octadecanoic acid     | C_{18}H_{36}O_{2} | 284 | 0.50 |
| 10 | 12.277   | Ergost-5-en-3-ol(3 beta) | C_{30}H_{48}O | 400 | 0.45 |
| 11 | 12.475   | Stigmasterol          | C_{28}H_{48}O | 412 | 0.32 |
| 12 | 13.387   | 22,23-dihydro Stigmasterol | C_{28}H_{46}O | 414 | 1.76 |
| 13 | 17.849   | Lanosta-8,24-dien-3-ol, acetate(3 beta) | C_{29}H_{46}O_{2} | 426 | 0.57 |

Table 2: Phytoconstituents of ethyl acetate extract of *Sphaeranthus indicus*

| No | RT (min) | Name of the Compounds | Molecular Formula | Molecular Weight | Height% |
|----|----------|-----------------------|------------------|-----------------|--------|
| 1  | 2.958    | Ethanol, 2-phenoxyc   | C_{2}H_{4}O_{2} | 138 | 0.30 |
| 2  | 2.995    | 2,4-Decadienol (E,E) | C_{16}H_{10}O | 152 | 9.54 |
| 3  | 3.068    | 1-Tetradecane        | C_{14}H_{28} | 196 | 0.47 |
| 4  | 3.138    | 1H-Benzopyran-2-one  | C_{15}H_{28}O | 160 | 0.80 |
| 5  | 3.338    | Phenol,2,4-bis(1,1-dimethylethyl) | C_{18}H_{20}O | 206 | 0.95 |
| 6  | 3.523    | 3-Octadecane         | C_{18}H_{36} | 252 | 4.07 |
| 7  | 3.675    | Hexadecanoic acid, methyl ester | C_{16}H_{32}O_{2} | 270 | 0.73 |
| 8  | 4.448    | n-Hexadecanoic acid  | C_{16}H_{32}O_{2} | 256 | 0.38 |
| 9  | 4.522    | 1-Hexacosanol        | C_{26}H_{50}O | 382 | 0.50 |
| 10 | 4.595    | Campesterol          | C_{20}H_{46}O | 400 | 0.45 |
| 11 | 4.642    | Lanosterol           | C_{29}H_{50}O | 426 | 0.32 |
| 12 | 4.863    | 9,19-Cyclolanost-24-en-3-ol (3 beta) | C_{30}H_{46}O | 426 | 1.76 |
| 13 | 4.988    | Lupeol               | C_{20}H_{30}O | 426 | 6.57 |

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