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RESEARCH ARTICLE

PHYTOCHEMICAL ANALYSIS OF LEAF EXTRACT OF PLANT *COSTUS SPICATUS* BY GCMS METHOD

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ABSTRACT

The bioactive components of *Costus spicatus* leaves have been evaluated using GC/MS. The chemical compositions of the ethanolic extract of *Costus spicatus* leaves were investigated using Perkin-Elmer Gas Chromatography–Mass Spectrometry, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) library. GC/MS analysis of ethanolic extract of *Costus spicatus* leaves revealed the existence of 1,2-Ethanediol, monoacetate (2.79%), 1-Tetradecanol (0.09%), Elema-1,3,11(13)-trine-12-ol (0.31%), beta-costol (0.33%), 4,7,10,13,16,19-Docosahexacnoic acid (0.57%), methyl ester (5.15%), Naphtho[1,2-6]furan-3-one (1.27%), 2,3,3a,4,5,5a,6,7,9a,9b- decahydro-3,5a-9-trimethyl-7,9a-peroxy (0.39%), Eremanthin (93.44%), 5-(ethynyl) nona-1,8-dien-5-ol (0.19%), Benzeneacetic acid (0.10%), Alpha-bergamotene (0.38%), 1,2-Benzenedicarboxylic acid, diisooctyl ester (2.55%), Isolongifolene, 4,5-dehydro- (3.68%) by GCMS analysis and the biological activity of each compound was discussed in this paper. As *Costus spicatus* can grow and spread easily and because of its higher biomass availability, it can prove as an effective and cheaper drug for various diseases.

Key words: *Costus spicatus* , GCMS analysis, Bioactive components.

1. INTRODUCTION

Phytochemistry or plant chemistry has developed in recent years as a distinct discipline, somewhere in between natural product organic chemistry and plant biochemistry and is closely related to both. It is concerned with the enormous variety of organic substances that are elaborated with and accumulated by plants and deals with the chemical structures of these substances, their biosynthesis, turn over and metabolism, their natural distribution and their biological function [1]. India is called the botanical garden of the world for its rich natural resources. Over 6,000 plants in India are used in traditional, folklore and herbal medicine. The Indian system of medicine has identified 1500 medicinal plants of which 500 are commonly used [2].

Phytochemicals are the chemicals extracted from plants. These organic chemicals are classified as primary or secondary constituents, depending on their role in plant metabolism. Primary constituents include the common sugars, aminoacids, proteins, purines and pyrimidines of nucleic acids, chlrophyll's etc. Secondary constituents are the remaining plant chemicals such as alkaloids (derived from aminoacids), terpenes (a group of lipids) and phenolics (derived from carbohydrates) [3]. Plant produces these chemicals to protect itself but recent research demonstrates that emphasizes the plant source of most of these protective, disease-preventing compounds. A true nutritional role for phytochemicals is becoming more probable every day as research uncovers more of their remarkable benefits [4].

Within a decade, there were a number of dramatic advances in analytical techniques including TLC, UV, NMR and GC-MS that were powerful tools for separation identification and structure determination of phytochemicals [5]. The aim of this study is to determine the organic compounds present in the *Costus spicatus* leaves extract with the aid of GC-MS Technique, which may provide an insight in its use in tradition medicine.

2. MATERIAL AND METHODS

2.1Collection of Plant materials:

Fresh plant material of *Costus spicatus* was collected from Kottyam District of Kerala and identified to confirm by the Taxonomist Botanical Survey of India, Tamilnadu, India.

2.2 Plant sample extraction

The leaves were cut into pieces and shade dried at room temperature. The dried leaves were subjected to size reduction to a coarse powder by using dry grinder and passed through sieve. 100 g of crushed leaves were continuously extracted with 95% ethanol using soxhlet up to 48 h. The extract was filtered and concentrated in rotatory evaporator at 35-40ºC under reduced pressure to obtain a semisolid material, which was then

lyophilized to get a powder (28.5%, w/v). Preliminary phytochemical tests were carried out on the ehtanolic extract of *Costus spicatus* leaves using standard procedures to identify the constituents as described by Malick and Singh, 1980[6], Segelman *et al.*, 1969[7] and Harborne [8,9].

2.3 GC –MS analysis

The GC-MS analysis was carried out using a Clarus 500 Perkin- Elmer (Auto System XL) Gas Chromatograph equipped and coupled to a mass detector Turbo mass gold – Perking Elmer Turbomas 5.2 spectrometer with an Elite-1 (100% Dimethyl ply siloxane), 300 m x 0.25 mm x 1 µm df capillary column. The instrument was set to an initial temperature of 110°C, and maintained at this temperature for 2 min. At the end of this period, the oven temperature was raised upto 280°C, at the rate of an increase of 5°C/min, and maintained for 9 min. Injection port temperature was ensured as 250°C and Helium flow rate as 1 ml/min. The ionization voltage was 70 eV. The samples were injected in split mode as 10:1. Mass Spectral scan range was set at 45-450 (mhz). The chemical constituents were identified by GC-MS. The fragmentation patterns of mass spectra were compared with those stored in the spectrometer database using National Institute of Standards and Technology Mass Spectral database (NIST-MS). The percentage of each component was calculated from relative peak area of each component in the chromatogram.

3. RESULTS AND DISCUSSION

Plants have an almost limitless ability to synthesize aromatic substances, most of which are phenols or their oxygen substituted derivatives. Most are secondary metabolites, of which at least 12,000 have been isolated, a number estimated to be less than 10% of the total. These substances serve as plant defense mechanisms against, insects and herbivores. Flavonoids exhibit several biological effects such as anti-inflammatory, anti-fungal, anti-hepatotoxic and anti-ulcer actions [10].

The phytochemical characters of the *Costus spicatus* leaves were investigated. The qualitative phytochemical analysis of ethanolic extract of *Costus spicatus* leaves extract contains alkaloids, flavonoids, glycosides, phenols, saponins, sterols and tannins which are an important in disease prevention and health preservation.

3.1 GC-MS ANALYSIS

The phytochemical compounds present in the ethanol extracts of *Costus spicatus* was identified by GCMS analysis. The active principles with their retention time (RT), molecular formula (MF) concentration (%) in the extract was presented. Totally fourteen compounds identified from the ethanol extract of the *Costus spicatus* are presented in Table 1. The plant sample relived the synthesis of 1,2-Ethanediol, monoacetate (2.79%), 1-Tetradecanol (0.09%), Elema-1,3,11(13)-trine-12-ol (0.31%), beta-costol (0.33%), 4,7,10,13,16,19-Docosahexacnoic acid (0.57%), methyl ester (5.15%), Naphtho[1,2-6]furan-3-one (1.27%), 2,3,3a,4,5,5a,6,7,9a,9b- decahydro-3,5a-9-trimethyl-7,9a-peroxy (0.39%), Eremanthin (93.44%), 5-(ethynyl) nona-1,8-dien-5-ol (0.19%), Benzeneacetic acid (0.10%), Alpha-bergamotene (0.38%), 1,2-Benzenedicarboxylic acid, diisooctyl ester (2.55%), Isolongifolene, 4,5-dehydro- (3.68%). The GC-MS chromatogram of ethanol extracts of *Costus spicatus* is shown in Figure 1. All these compounds are of pharmacological importance as they possess the properties such as anti-diabetic, analgesic, antibacterial, and antifungal activity.

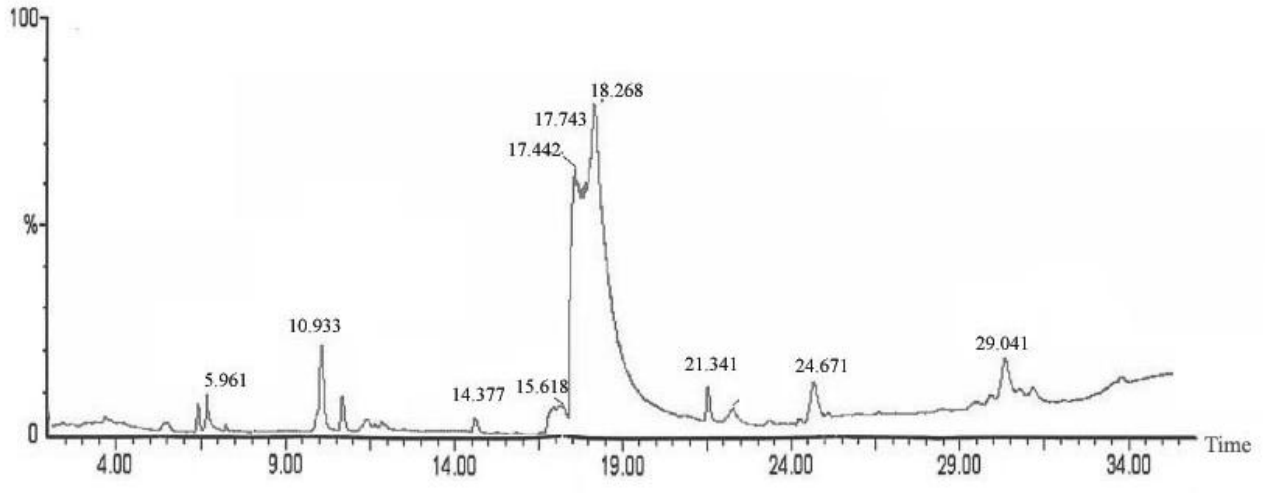

Figure 1: Chromatogram obtained from the GCMS with the extract of *Costus spicatus* leaves

Table 1 shows the components identified in ethanolic extract of *Costus spicatus* leaves (GC MS method)

S. No	RT	Compound	Molecular formula	Relative content (%)
1	5.961	1,2-Ethanediol, monoacetate	$C_4H_8O_3$	2.79%
2	10.933	1-Tetradecanol	$C_{14}H_{30}O$	0.09%
3	14.377	(-)-Elema-1,3,11(13)-trine-12-ol	$C_{11}H_{12}O_3$	0.31%
4	15.618	(+)-beta-costol	$C_{15}H_{24}O$	0.33%
5	17.442	4,7,10,13,16,19-Docosahexacnoic acid	$C_{25}H_{38}O_2$	0.57%
6	17.743	methyl ester	$C_{13}H_{26}O_2$	5.15%
7	17.960	Naphtho[1,2-6]furan-3-one	$C_{26}H_{30}O_8$	1.27%
8	18.021	2,3,3a,4,5,5a,6,7,9a,9b-decahydro-3,5a-9-trimethyl-7,9a-peroxy	$C_{20}H_{28}O_3$	0.39%
9	18.236	Eremanthin	$C_{17}H_{25}NO_2$	93.44%
10	21.341	5-(ethynyl) nona-1,8-dien-5-ol	$C_{21}H_{30}O_2$	0.19%
11	22.125	Benzeneacetic acid	$C_8H_8O_2$	0.10%
12	23.954	Alpha-bergamotene	$C_{15}H_{24}$	0.38%
13	24.671	1,2-Benzenedicarboxylic acid, diisooctyl ester	$C_{24}H_{38}O_4$	2.55%
14	29.041	Isolongifolene, 4,5-dehydro-	$C_{15}H_{22}$	3.68%

In the present study fourteen chemical constituents have been identified from ethanolic extract of the plant of *Costus spicatus* by Gas Chromatogram- Mass spectrometry (GCMS) analysis. Thus each compound identified in leaf extract of Costus spicatus has its own biological importance and further study of this plant's phytochemical by insilico and invitro methods can prove its medicinal importance in future and can be an effective and efficient drug source in cheaper rate as it has higher biomass availability.

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