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GC-MS DETERMINATION OF BIOACTIVE COMPONENTS OF PIPER NIGRUM

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ABSTRACT

In this study, a gas chromatography – mass spectrometry method is successfully developed for the determination of chemical components which were present in black pepper *Piper nigrum.* The samples were extracted with petroleum ether, cleaned up and purified through solid phase extraction. The present investigation deals with GC-MS analysis of petroleum ether extract of *Piper nigrum.* The petroleum ether extract of *Piper nigrum* shown the GC-MS analysis revealed the twenty two compounds were identified.

KEYWORDS: GC-MS analysis, *Piper nigrum.*

INTRODUCTION

Black pepper *Piper nigrum. L* is a flowering vine in the family of Piperaceae cultivated for its fruit which is usually dried and used as a spice and seasoning. *P.nigrum* is one of the oldest and most widely used spices in the world. It is produced from the unripe but fully developed fruits. *P.nigrum* has the characteristics of pepper aroma and flavor due to their chemical substances. Black pepper is native to South East Asia and is extensively cultivated in tropical regions (Nahak *et al.,* 2011). The main pungent principle in the green berries of *P.nigrum* is piperine. Atleast five other alkaloids structurally related to the piperine also occur in smaller amounts. The extracts from black pepper have been found to have anti-oxidant properties and anti-carcinogenic effects especially when compared to chilli (Nalini *et al.,* 2006). Cardamom, clove and pepper are some of the commonly used spices in the every Indian household for culinary purposes as well as in the treatment of various infections (Mittal *et al.,* 2014).

Pepper is also used in folk medicine as aphrodisiac, carminative, stomachic, antiseptic diuretic and for the treatment of cough, rheumatoid arthritis, peripheral neuropathy, melanoderma and leprosy due to the presence of volatile compounds, tannins, phenols and other unknown substances. However, many researches have been carried out on Black pepper *Piper nigrum. L* in India. Some disagreement about the presence of its constituents was observed. Therefore, present work was undertaken to carry out a complete investigation of the compounds of *P.nigrum* with GC-MS analysis.

MATERIALS AND METHODS

Collection of plant material

The fresh black pepper was obtained from the local market of pollachi city, Coimbatore district, Tamilnadu, India. The pepper was dried and ground by using mixer grinder. Then the powder was sieved prior to the extraction process.

Preparation of samples

Piper nigrum powder was macerated in petroleum ether for 10 days by occasional stirring. After ten days the extract was filtered using Whatman no.1filter paper. The residue obtained after the filteration of petroleum ether extract of *P.nigrum* was again extracted by Soxhlation method. The extract was taken and filtered. The obtained extract was concentrated by rotary evaporator at 60° C and these were analyzed by gas chromatography – mass spectrometry (GCMS).

Identification of components

Interpretation of mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular formula, molecular weight, area(%), structure of the components of the test materials were ascertained.

RESULT

The identified compounds and their retention time, molecular formula, molecular weight, peak area (%) and activities related with medicinal uses are given in Table.1

Table 1: Quantitative Constituents of Petroleum Ether Extract of *Piper nigrum*.

S. No.	Retention Time (RT)	Area%	Name	Molecular weight	Molecular formula
1	19.059	2.70	α- Cubebene	204	$C_{15}H_{24}$
2	19.437	0.91	Aromadendrene	204	$C_{15}H_{24}$
3	20.273	26.98	Caryophyllene	204	$C_{15}H_{24}$
4	20.492	0.38	Cyclopropane	226	$C_{12}H_{18}O_2S$
5	20.992	0.19	1,6,10-Dodecatriene	204	$C_{15}H_{24}$
6	21.128	2.64	1,4,8-Cycloundecatriene	204	$C_{15}H_{24}$
7	21.642	0.12	3,5- Dodecadiyne	176	$C_{13}H_{20}$
8	21.812	0.33	1,6 –Cyclodecadiene	204	$C_{15}H_{24}$
9	21.962	1.93	Naphthalene	204	$C_{15}H_{24}$
10	22.179	3.47	Azulene	204	$C_{15}H_{24}$
11	22.364	9.96	Cyclohexene	204	$C_{15}H_{24}$
12	22.647	4.54	Naphthalene	204	$C_{15}H_{24}$
13	23.640	2.26	2,6,10-Dodecatriene-1-ol	222	$C_{15}H_{26}O$
14	24.185	0.34	Penta decadiynoic acid	234	$C_{15}H_{22}O_2$
15	24.353	3.18	Androstan-17-one	318	$C_{21}H_{34}O_2$
16	24.575	0.14	Octadecadiyonic acid	204	$C_{13}H_{20}N_2$
17	24.823	0.17	Cyclopropane	204	$C_{13}H_{20}N_2$
18	24.962	0.28	Methanoazulene	206	$C_{15}H_{26}$
19	25.336	0.71	β-Vatirenene	202	$C_{15}H_{22}$
20	25.906	2.42	Methyl octadec-9-en-12-ynoate	204	$C_{15}H_{24}$
21	33.296	0.17	Tetradecanal	204	$C_{15}H_{24}$
22	35.311	0.34	Oxirane	212	$C_{14}H_{28}O$

Analysis of the chemical composition of the extract by GC-MS facilitated the identification of components in petroleum ether extract of *Piper nigrum* were α-Cubebene(2.70%), Aromadendrene (0.91%), Caryophyllene (26.98%), Cyclopropane (0.38%), 1,6,10-Dodecatriene (0.19%),1,4,8-Cycloundecatriene (2.64%), 3,5- Dodecadiyne (0.12%), 1,6 –Cyclodecadiene (0.33%), Naphthalene (1.93%), Azulene (3.47%), Cyclohexene (99.96%), Penta decadiynoic acid (0.34%), Androstan-17-one(3.18%), Octadecadiyonic acid (0.14%), Cyclopropane (0.17%), Methanoazulene (0.28%), β-Vatirenene (0.71%), Methyl octadec-9-en-12-ynoate (2.42%), Tetradecanal(0.17%) and Oxirane (0.34%).(Table. 1).

Chromatogrampep291216aftD:\pep291216aft\sample. qgd

Intensity

Chromatogram pep291216 aft D:\pep291216 aft\sample.qgd
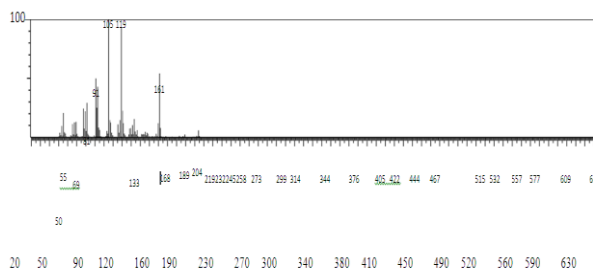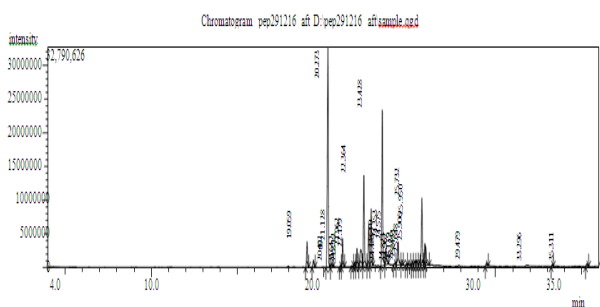
Hit#:1 Entry:110121 Library:WILEY8.LIB
SI:94 Formula:C15H24 CAS:3856-25-5
MolWeight:204 RetIndex:0
CompName: TRICYCLO[4.4.0.0(2,7)]DEC-3-ENE, 1,3-DIMETHYL-8-(1-METHYLETHYL)-, ST $$ TRICYCLO[4.4.0.0(2,7)]DEC-3-ENE, 1,
TIC*1.00

DISCUSSION

The gas chromatogram shows that the relative concentrations of various compounds are getting eluted as a function of retention time (RT). The height of the peaks indicates the relative concentrations of the compounds present in the plant. The mass spectrometer analyzes of the compound eluted at different times to identify the nature and structure of the compounds. The large compound fragments into small compounds give rise to appearance of peaks at different m/z ratios. These mass spectra are fingerprint of that compound which can be identified from the data library.

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