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GAS CHROMATOGRAPHY-MASS SPECTROSCOPIC ANALYSIS OF BLACK PLUM SEED (SYZYGIUM CUMINI) EXTRACT IN HEXANE

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ABSTRACT

Objective: Black plum seed has unique medicinal value and used as antidiabetics, antibacterial, and anti-inflammatory.

Methods: Gas chromatography-mass spectrometry (GC-MS) analysis of the extracts of black plum seed obtained using solvent extraction with hexane as a solvent is used to attempt the identification of prominent components.

Results: Black plum seed extract is obtained by solvent extraction technique using Soxhlet extractor. A total of 10 compounds are predicted in black plum seed extract by GC-MS analysis.

Conclusion: The work presented relates to the study of GC-MS analysis of the extract of black plum seed obtained using solvent extraction with hexane as a solvent. Of ten compounds of black plum seed extract, five compounds are known to have antimicrobial properties.

Keywords: Gas chromatography-mass spectrometry, Black plum, Syzygium cumini, Seed extract.

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INTRODUCTION

Black plum's scientific name is *Syzygium cumini*, in India, and it is commonly known as Jamun. The tree is a commonly present in India and other tropical and subtropical regions of the world. Black plum tree has a great economic value because most of the parts of the tree such as the fruit, leaves, bark, and seeds are used as medicine to treat various diseases. For example, it is used to manage the blood sugar level in the patients suffering from diabetes. There are reports of study conducted to study the effects of various concentrations (0.0, 1.56, 3.125, 6.25, 12.5, 25, 50, and 100 μ g/ml) of the leaf extract of *S. cumini* Linn or *Eugenia cumini* (black plum, Jamun, family: Myrtaceae) on the alteration in the radiation-induced micronuclei formation in the cultured human peripheral blood lymphocytes [1].

Analysis of the antioxidant activity of the fruit skin using different assays, such as hydroxyl radical-scavenging assay, based on the benzoic acid hydroxylation method, superoxide radical-scavenging assay, based on photochemical reduction of nitro blue tetrazolium (NBT) in the presence of a riboflavin-light-NBT system, 2,2-diphenyl-1-picrylhydrazyl radicalscavenging assay, and lipid peroxidation assay using egg yolk as the lipid-rich source confirms the antioxidant activity [2]. Characterized and evaluation of anthocyanin pigments from S. cumini fruit peels for their antioxidant efficacy, and stability as extract and in formulation is also reported [3], and the study identified three anthocyanins as gluco glucosides of delphinidin, petunidin, and malvidin by high-performance liquid chromatography (HPLC)-ESI-MS. Gas chromatography-mass spectrometry (GC-MS) technique is one of the effective techniques in a study related to medicinal properties and antibacterial properties of herbal extracts [4]. For the extraction of volatile components from the black plum fruits by simultaneous distillation and solvent extraction method, modified Likens and Nickerson apparatus can be used, and the analysis of volatile compounds by GC-MS showed the presence of 30 compounds [5]. There are studies conducted to deal with the downstream processing of anthocyanins from black plum

(*S. cumini* L.) to obtain anthocyanins in a purified form [6], this study includes adsorption employing six different adsorbents, and among these, Amberlite XAD7HP showed the highest adsorption capacity (1.07 mg/mL of adsorbent) and desorption ratio (87.62%).

Chemoprotective activity of black plum seed extract against *in vivo* oxidative stress and genomic is also established [7]. There are studies, showing that black plum seed extract can possibly play a vital role as a chemopreventive agent against oxidative stress and genomic damage [7]. Black plum seed kernel extracts exhibit the inhibition of α -glucosidase from mammalian (rat intestine), bacterial (*Bacillus stearothermophilus*), and yeast (*Saccharomyces cerevisiae*, Baker's yeast). The *in vitro* studies using the mammalian α -glucosidase from rat intestine indicated the extracts to be more effective in inhibiting maltase when compared to the acarbose control [8], and other techniques such as HPLC and nuclear magnetic resonance are also useful in the identification of constituents of extract [9].

In recent times, we used supercritical fluid extraction technology (instrument name: SFC L-tex Japan) to extract the compounds from various biological materials such as plants and animals, performed GC-MS analysis of oil extracted for freshwater crab, and also analyzed fatty acid composition of some animals [10-16].

METHODS

Black plum seed powder is purchased from local market of Aurangabad city, Maharashtra, India. Black plum seed powder is subjected to Soxhlet extraction process using hexane as a solvent to obtain black palm seed extract.

GC-MS is a unique method for the analysis and measures the quantity of organic volatile and semi-volatile compounds. GS is employed to separate mixtures into individual components employing a temperature-controlled capillary column. MS is applied to recognize a



Fig. 1: Gas chromatography mass spectrometry analysis spectrum of black plum seed extract

Table 1: Specification of GC-MS

Conditions during GC-MS analysis	
Runtime (min)	54.09
Injection volume(µl)	1.00
Scans	6439
Low mass (m/z)	40
High mass (m/z)	400
Gas	Helium
Solvent	Hexane

GC-MS: Gas chromatography-mass spectrometry

variety of components from their mass spectra. In the present study, black plum seed powder is used for extraction and analysis to study the constituents. Black plum seed extract is extracted by solvent extraction technique using Soxhlet extractor with hexane as a solvent. The GC-MS analysis is carried out at SAIF, Chandigarh University. The operating conditions used in the GC-MS analysis are given in Table 1.

RESULTS AND DISCUSSION

The GC-MS chromatography of black plum seed extract shows different peaks (Fig. 1). Each peak is representing a constituent present in the extract. These peaks are further analyzed and the fractions. So obtained, at different retention time is characterized by MS, which is represented in Fig. 2.

GC-MS analysis of black plum seed extract reveals that the seed contains 10 different compounds. Table 2 shows that, among all compounds, compound with retention time 20.33 shows highest concentration (39.98 %), followed by compound with retention time 14.42 (30.28 %), compound with retention time 12.39 (20.30 %), compound with retention time 11.29 (2.61%), compound with retention time 10.95 (1.63%), compound with retention time 11.34 (1.49%), and compound with retention time 11.94 (1.45%), and concentrations of remaining compounds are <1%. These are probable compounds based on GC-MS compound library search.

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Compound name	Area %	Molecular formula	Molecular weight	RT (min)	Peak area	Structure
Oleic acid	30.28	C ₁₈ H ₃₄ O ₂	282	14.42	155781647.79	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~
n-hexadecanoic acid	20.30	$C_{16}H_{32}O_{2}$	256	12.39	104407935.16	H0
Cyclooctasiloxane, hexadecamethyl	0.79	$C_{16}H_{48}O_8Si_8$	593.2315	10.04	4076063.86	- CANA
1-monolinoleoylglycerol trimethylsilyl ether	1.45	$C_{27}H_{54}O_4Si_2$	498	11.94	7461639.22	<
Octadecanal, 2-bromo- Cyclohexasiloxane, dodecamethyl-	2.61 0.79	$\begin{array}{c} C_{18}H_{35}BrO\\ C_{12}H_{36}O_6Si_6 \end{array}$	346 444.924	11.29 7.24	13411130.27 4081565.85	
Cycloheptasiloxane, tetradecamethyl-	0.69	$C_{14}H_{42}O_{7}Si_{7}$	519.078	8.72	3531066.64	- Jan Ala
Pyrazole[4,5-b] imidazole, 1-formyl-3-ethyl-6-á-d-ribofuranosyl-	1.63	$C_{12}H_{16}N_4O_5$	296.283	10.95	8380945.03	
Stearic acid, 3-(octadecyloxy) propyl ester Benzaldehyde, 2,4,5-trimethoxy-	1.49 39.98	$\begin{array}{c} C_{39}H_{78}O_{3}\\ C_{10}H_{12}O_{4} \end{array}$	595.05 196.2	11.34 20.33	7641092.59 205642953.61	or of the second



Fig. 2: Prominent peaks of gas chromatography-mass spectrometry of black plum seed extract, (a) spectrum with retention time = 7.24,
(b) spectrum with retention time = 8.72, (c) spectrum with retention time = 10.04, (d) spectrum with retention time = 10.95, (e) spectrum with retention time = 11.29, (f) spectrum with retention time = 11.34, (g) spectrum with retention time = 11.94, (h) spectrum with retention time = 12.39, (i) spectrum with retention time = 14.42, (j) spectrum with retention time = 20.33

S. no	Compound name	Reported bioactivity
1	Oleic acid	Antimicrobial
2	n-hexadecanoic acid	Anti-inflammatory, Antioxidant, hypocholesterolemic nematicide, pesticide, anti-androgenic
		flavor, hemolytic, 5-Alpha reductase inhibitor, potent mosquito larvicide
3	Cyclooctasiloxane, hexadecamethyl	Conditioning agent
4	1-Monolinoleoylglycerol trimethylsilyl ether	Antimicrobial Antioxidant, Anti-inflammatory antiarthritic, antiasthma, diuretic
5	Octadecanal, 2-bromo-	Nontoxic and efficient anti-microbial agents

Black plum seed extract contains 10 different compounds. From these ten compounds, 5 compounds possess antimicrobial properties. Oleic acid possesses antimicrobial property [17]. n-hexadecanoic acid possesses following properties: Anti-inflammatory, antioxidant, hypocholesterolemic nematicide, pesticide, anti-androgenic flavor, hemolytic, 5-alpha-reductase inhibitor, and potent mosquito larvicide [17]. Cyclooctasiloxane and hexadecamethyl are used as conditioning agent [18]. 1-monolinoleoylglycerol trimethylsilyl ether possesses following properties: Antimicrobial, antioxidant, antiinflammatory, antiarthritic, antiasthma, and diuretic [19]. Octadecanal, 2-bromo- is non-toxic and efficient antimicrobial agent [20].

CONCLUSION

The work presented relates to the study of GC-MS analysis of the extracts of black plum seed obtained using solvent extraction with hexane as a solvent. The extract is found to contain many medicinally active compounds. Ten compounds are identified and details presented, five of which are found to exhibit antimicrobial activity against different diseases. The medicinally active compounds can be isolated and considered for the preparation of medicine.

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AUTHORS' CONTRIBUTIONS

All authors have equally contributed at all stages of the work except extraction; extraction was done by Abdul Jaleel A. H under the

supervision of Dr. Yusuf Haneef Shaikh. Analysis of chromatography report was done with the help of Dr. Mazhar Farooqui.

CONFLICTS OF INTEREST

The author hereby declares no conflict of interest regarding the manuscript and experimentation done.

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