

Analysis of lipid metabolites through GCMS-FID from papaya seed oil for detecting potential uses for therapeutic and nutritional

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Abstract

Oil extract through hexane solvents of the *Carica papaya* seeds were used for detection of metabolites. The experiment was designed at Junagadh Agricultural University to analyze the metabolic profiles of papaya seeds in order to high lighten the important phytoconstituents presents in seeds. Mostly the untargeted metabolite fingerprinting was done by using GC-MS Instruments and software for detection of similar compounds and structurer NIST library were used. Generated compound data were compared with electronic database search Google and electronic library search for articles published in peer-reviewed journals and uploaded of fatty acids profiles. Analyzed through GCMS data displayed twenty-nine peaks of metabolites in the papaya seed extracts. The data includes RT, Area % and class of compounds. The results shown mostly were fatty acids out of them the concentration of four fatty acids were higher in seeds namely 9-Octadecenoic acid i.e oleic acids (17.83 %) Hexadecanoic acid (4.31 %) Octadecanoic acid (1.69%), 17.17 % area with beta sitosterol's, 9,12-Octadecadienoic acid (2.65) while remaining were found to be less than 1 % area. The 1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester were detected at RT of 19.01 with 0.16% area and it was proven as anticancer metabolites. Many of compounds have properties of pharmaceuticals and food industries.

Keywords: GC MS, papaya, fatty acids

Introduction

Papaya (*Carica papaya* L.) is one of the important tropical and subtropical fruit in the world, originated in Mexico as a cross between two species of the genus *Carica*. India is the largest producer of papaya in the world but productivity is lower than the other countries like Brazil, Indonesia and Mexico. One of the main reasons is the absence of high yielding, disease resistant hybrid varieties. (Manjunath *et al.* 2024) ^[3] The Indian papaya have many uses other then fruit crops. It also used in pammasal as "tootifrutti" or dice or slice with coloured sugary material and slices also as ingredients for making fast cooking process in dahl. and many of these traditional uses and reported anti-viral, anti-bacterial, anti-protozoal, anti-fungal, anti-inflammatory, anti-tumour, anti-hypertensive, wound-healing, neuroprotective, diuretic, abortifacient, anti-fertility, hypoglycaemic, and hypolipidemic properties. Papaya can be made into jam, jelly, nectar, dried into slabs, canned in the form of slice and the fruit powder. (Parmar *et al.* 2020) ^[4]. The papaya plant parts (seeds, roots, leaves, barks, flowers, latex, and fruit) have been used in folk medicine to treat various diseases, including diabetes, cancer, and cardiovascular and infectious diseases (Li H *et al.* 2023) ^[2]. Therefore, the waste products as seeds can be specialized parts and it has tremendous possibility to dig out the compounds in especially in seed parts of papaya. As one of the reasons behind to study on seed oil of papaya.

Lipid metabolites play a crucial role in various biological processes, including cellular signalling, energy storage, and membrane structure. In recent years, there has been a growing interest in exploring the potential health benefits of plant-derived oils due to their rich and diverse lipid profiles. Among these, papaya seed oil, a byproduct of the papaya fruit (*Carica papaya*), has gained attention for its unique

composition and potential therapeutic and nutritional benefits. Papaya seeds are typically discarded as waste, yet they are a valuable source of bioactive compounds, including essential fatty acids, antioxidants, and other lipid metabolites. These compounds have been linked to various health benefits, such as anti-inflammatory, antimicrobial, and antioxidant properties. Despite this, the detailed characterization of lipid metabolites in papaya seed oil remains relatively unexplored, particularly using advanced analytical techniques. Gas Chromatography-Mass Spectrometry with Flame Ionization Detection (GC-MS-FID) is a powerful analytical method for the detailed profiling of lipid metabolites. This technique allows for the separation, identification, and quantification of various fatty acids and lipid components with high precision. By applying GC-MS-FID to papaya seed oil, the generated information can give the comprehensive understanding of its lipid composition, uncovering potential bioactive compounds that could be utilized in therapeutic and nutritional applications. The objective of this study was to analyse the lipid metabolites in papaya seed oil using GC-MS-FID to identify key components and assess their potential applications. This research aims to provide insights into the nutritional value and therapeutic potential of papaya seed oil, contributing to its broader application in health and wellness sectors. The findings can also support the development of new functional foods and therapeutic products, highlighting the value of papaya seed oil beyond its traditional use as agricultural waste.

Materials and Methods

The standard condition and protocol for Gas Chromatography-Mass Spectrometry with Flame Ionization Detection (GC-MS-FID) was used for detailed profiling of

lipid metabolites from oils of Papaya seeds with some modification and protocol developed at FTL JAU Junagadh with some modification (Umamaheshwar *et al.*2026) ^[14]

GCMS conditions: Extracted oils was used for derivatives and make methyl ester and applied in autosampler tubes. GC-MS analyses were performed using a DB-5MS capillary column (30 m × 0.25 mm i.d., film thickness 0.25 µm, Agilent Technologies, Santa Clara, CA, USA). The chromatographic runs were started as per the following details conditions [GC-2010]. The GCMS conditions as per table indicated: ionization mode, electron ionization (EI) mode; ionization current, 60 µA; ionization voltage, 70 eV. Each sample was analysed. Eluted compounds were identified using the NIST (<http://www.nist.gov/srd/nist1a.cfm>), the Wiley NBS, the mass spectral data as well as used pubmed and other data sources for detection. Fatty acids and compounds were used for google search, NIST library for the potential application for food and industrial application.

Instruments protocol for analysis of GCMS-FAME

Column Oven Temp.	40.0 °C	MS table: Start Time :3.50min, End Time :54.67min, ACQ Mode: Scan, Event Time :0.50sec, Scan Speed 1428, Start m/z :50.00, End m/z :700.00, Sample Inlet Unit :GC "GCMS-QP2010 Plus], Ion Source Temp :200.00 °C, Interface Temp.:290.00 °C, Solvent Cut Time :2.00 min, Detector Gain Mode: Relative Detector Gain: +0.00 kV, Threshold 100"
Injection Temp.	:250.00 °C	
Injection Mode	: Split	
Flow Control Mode	: Pressure	
Pressure	:49.5 kPa	
Total Flow	:54.0 mL/min	
Column Flow	:1.00 mL/min	
Linear Velocity	:36.1 cm/sec	
Purge Flow	:3.0 mL/min	
Split Ratio	:50.0	
High Pressure Injection	: ON	
High Press. Inj. Pressure	:250.0 kPa	
High Press. Inj. Time	:1.00 min	
Carrier Gas Saver	: OFF	
Splitter Hold	: OFF	
Oven Temp. Program		
Rate	Temperature(°C)	Hold Time(min)
-	40.0	3.00
6.00	290.0	10.00

Results and Discussion

The results for lipid content in papaya seed oils with the area % and compounds detected are presented in Table.1 and their potential uses are presented in table.2. The results revealed 27 compounds in oils out of them 13 compounds were detected as fatty acids and their derivatives viz, peak no. 3,6,7,10,11,12,13,15,21,22,27,28 and29. The putative fatty acids were detected at RT from 11.465 to 29.511 minutes. The distribution pattern of fatty acids and lipids compounds showed more than one percentage area includes the compounds viz., (4.31%) Hexadecanoic acid, (1.69 %) Octadecanoic acid, (17.83 %) Oleic acids, (2.65 %) Linoleic acids, (3.95%)n-Hexadecanoic acid, (17.17%) beta.-Sitosterol, (35.72%) Cholestane-3,5-diol, 5-acetate, (3.beta.,5.alpha.), (1.33 %) Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester,(3.24 %) 9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E,E,E)-, (1.43 %) Stearic acids and (6.71 %) Oleic Acid. These results are in line with Yanty *et al.* (2014) ^[6] ^[6] who studied Papaya seed oil and they found ten detectable fatty acids, of which 78.33% were unsaturated. Oleic (73.5%) acid was the dominant fatty acids followed by palmitic acid (15.8%). Based on the high-performance liquid chromatography (HPLC) analysis. Out of total 29 compounds in papaya seed four compounds was found to be 77.43% area. means remaining were less than 5 %. While maximum area was detected for steroids in papaya dry seeds with 35.72 % as Cholestane-3,5-diol, 5-acetate, (3. beta.,5. alpha.) and next to this 17.83% at RT of 17.194 fatty acids was detected. 1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester were found in all four horticulture crop seed oils. (Save *et al.* 2015) ^[5], ^[5] 11,14,17-Eicosatrienoic acid, fatty acid only found in sapota. Similarly for 17.17 % area with beta sitosterols only found in papaya, Beta-sitosterol blocks cholesterol absorption and has been shown in studies to reduce blood levels of cholesterol, and 35.72 % of Cholestane-3,5-diol, 5-acetate, (3.beta.,5.alpha.)- were detected as sterols this results are in agreement with the qualitative and quantitative chemical compositions of composition of seed oils from *Carica papaya* L. fruits, cultivated in Russia and the seed oils were determined by a combination of mass spectrometry and NMR spectroscopy. By Goriainov *et al.* (2023) ^[1], ^[1] they found content of sterols and triterpene alcohols was (537.5–918.2) mg/100 g of oil (0.54–0.92%), and up to 75% of the fraction was represented by β-sitosterol (55.9–66.7%) and its saturated analogue-sitostanol (11.0–15.7%). The physicochemical properties and the fatty acid, sterol, and triterpene alcohol

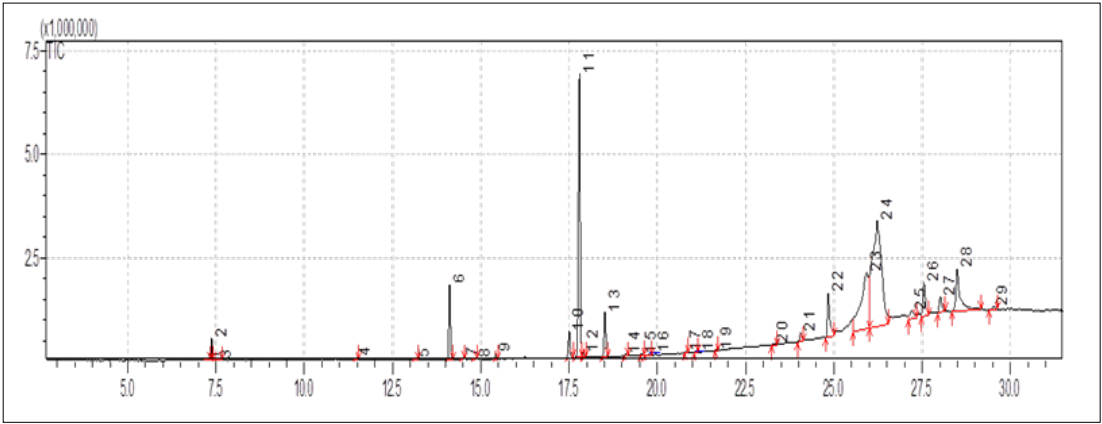


Fig 1: Chromatogram of Papaya seed oil through the GC MS

Table 1: Lipid derived metabolites from papaya seed oils.

Peak No	RT (min)	Area (%)	Class	Compound Identified
1	7.175	0.08	Odour flavour	Butanoic acid, 2-methyl-, heptyl ester
2	7.378	0.74	Aromatic hydrocarbon	Naphthalene
3	7.524	0.19	Fatty alcohol esters.	Formic acid, octyl ester
4	11.465	0.13	saturated fatty acid	Octanoic Acid
5	13.135	0.1	saturated fatty acid	Nonanoic acid
6	14.115	4.31	Fatty acids	Hexadecanoic acid
7	14.492	0.11	Fatty acids	6-Octadecenoic acid,
8	14.84	0.1	Acid dimer	trans-2-undecenoic acid
9	15.422	0.29	Phenols	Phenol, 2,4-bis(1,1-dimethylethyl)-
10	17.501	1.69	Fatty acids	Octadecanoic acid
11	17.794	17.83	Fatty acids	9-Octadecenoic acid (Z) (Oleic acids)
12	17.9	0.28	Fatty acids	9-Octadecenoic acid (Z)-, methyl ester
13	18.515	2.65	Fatty acids	9,12-Octadecadienoic acid (Linoleic acids)
14	19.101	0.16		1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester
15	19.557	0.14	Fatty acids	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-
16	19.783	0.15	Flavour of seeds	Methyl 3-(3,5-di-tert-butyl-4-hydroxyphenyl)propionate
17	20.813	0.1	saturated fatty acid	Heneicosanoic acid
18	21.094	0.1	ethylene sulfide	Thiirane, octyl-
19	21.681	0.09	D-glucose	3-O-Methyl-d-glucose
20	23.292	0.14	nine-carbon fatty acid	Nonanoic acid
21	24.06	0.67	fatty acid deri.	Hexadecanoic acid, 2-hydroxy-1,3-propanediyl ester
22	24.842	3.95	Fattyacids	n-Hexadecanoic acid
23	25.935	17.17	phytosterols	beta.-Sitosterol
24	26.229	35.72	Steroids	Cholestane-3,5-diol, 5-acetate, (3.beta.,5.alpha.)-
25	27.198	1.33	1,2-Dipalmitoylglycero	Hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester
26	27.55	3.24	Triolein	9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E,E,E)-
27	28.014	1.43	Fatty acids	Octadecanoic acid (Stearic acids)
28	28.492	6.71	Fatty acids	Oleic Acid
29	29.511	0.4	Fatty acids	9,12-Octadecadienoic acid

Table 2: Industrial and therapeutic uses of metabolites detected in seed oils of papaya

Chemical Name	Used in	Reference
Butanoic acid, 2-methyl-, heptyl ester	Flavourings and fragrances, potentially in pharmaceuticals for taste masking.	(Pino and Queris, 2012) [13], [13]
1,2-Benzene (Catechol / Benzene derivative)	Antiseptics and to treat insect bites, but its use is limited due to potential toxicity.	(Pino and Queris, 2012) [13], [13]
Octyl formate	Fragrances and as a solvent, with potential applications in pharmaceutical formulations for controlled release.	(Pino and Queris, 2012) [13], [13]
Caprylic acid	Anti-fungal treatments and as a medium-chain triglyceride in nutritional supplements.	(Pino and Queris, 2012) [13], [13]
Pelargonic acid	As anti-fungal and antimicrobial agents and as a flavouring agent.	(Pino and Queris, 2012) [13], [13]
Palmitic acid	Skincare products and supplements due to its role in lipid metabolism and cell membrane integrity.	(Al-Asadi <i>et al.</i> , 2021) [7], [7] (Kabore <i>et al.</i> , 2023) [9], [9]
Oleic acid	Skincare for its emollient properties and in dietary supplements for cardiovascular health.	(Al-Asadi <i>et al.</i> , 2021) [7], [7]
Undecenoic acid	Anti-Fungal treatments and as a component in some skin care products.	(Oloyede <i>et al.</i> , 2019) [12], [12]
2,4-Bis(tert-butyl)phenol	As antioxidant in pharmaceuticals and cosmetics.	(Malathi and Sujitha, 2025) [10], [10]
Stearic acid	As a lubricant and in cosmetics for its moisturizing properties.	(Kabore <i>et al.</i> , 2023) [9], [9]
Oleic acid	Emollient and anti-inflammatory properties in skincare and supplements.	(Nayan <i>et al.</i> , 2017) [11], [11]
Methyl oleate	To improve drug solubility.	(Ezekwe and Chikezie, 2017) [8], [8]
Linoleic acid	Dermatology for its role in skin barrier function and in dietary supplements.	(Malathi and Sujitha, 2025) [10], [10]
Diisobutyl phthalate	As a plasticizer in medical devices.	(Malathi and Sujitha, 2025) [10], [10]
Methyl alpha-linolenate	Dietary supplements for cardiovascular and anti-inflammatory benefits.	(Al-Asadi <i>et al.</i> , 2021) [7], [7]
Methyl 3-(3,5-di-tert-butyl-4-hydroxyphenyl)propionate	Pharmaceutical and cosmetic formulations.	(Malathi and Sujitha, 2025) [10], [10]
Heneicosanoic acid	pharmaceutical and cosmetic formulations	(Al-Asadi <i>et al.</i> , 2021) [7], [7]
Octyl thiirane	In specialized chemical synthesis.	(Pino and Queris, 2012) [13]
3-O-Methyl-D-glucose	in biochemical research	(Ezekwe and Chikezie, 2017) [8], [8]
Palmitic acid, glycerol ester	Moisturizing and emollient properties.	(Kabore <i>et al.</i> , 2023) [9], [9]
Beta-sitosterol	Used in cholesterol-lowering supplements and as an anti-inflammatory agent.	(Al-Asadi <i>et al.</i> , 2021) [7], [7]

Cholestane-3,5-diol 5-acetate	Studied for its potential role in cholesterol metabolism and related health conditions.	(Malathi and Sujitha, 2025) ^{[10], [10]}
Triolein	Used in food and pharmaceutical formulations for its emulsifying properties.	(Kabore <i>et al.</i> , 2023) ^{[9], [9]}

Conclusions

Hexane extracts of *Carica papaya* seed oils were used for metabolites detection and FAME using were used for fingerprinting through GC-MS instruments and their software for detecting related compounds by using NIST library. and The compound data were compared with generated E- data base for pharmaceutical and uses. GCMS analysis revealed twenty-nine metabolite peaks and the majority of these were of lipid class, out of them eleven had higher % area. Specifically, Stroides and beta Sitosterol both constitutes more than 50% of all metabolites, 9-Octadecenoic acid (Z) (17.83%), Hexadecanoic acid (4.31%), Octadecanoic acid (1.69%), and 9,12-Octadecadienoic acid (2.65). The remaining 25 chemicals accounting for less than 1% of the total area. The 1,2-Benzenedicarboxylic acid bis(2-methylpropyl) ester was detected at RT of 19.01 with 0.16 percent area, and it has anticancer characteristics in a variety of plants. The study revealed that many compounds have medicinal value in seed oils of *Carica papaya*.

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