OPEN ACCESS

Scholars International Journal of Biochemistry

Abbreviated Key Title: Sch Int J Biochem
ISSN 2616-8650 (Print) | ISSN 2617-3476 (Online)
Scholars Middle East Publishers, Dubai, United Arab Emirates
Journal homepage: https://saudijournals.com

Original Research Article

GC-MS Analysis of Methanol Seeds Extract of *Persea americana*

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DOI: https://doi.org/10.36348/sijb.2026.v09i03.001 | **Received:** 29.08.2026 | **Accepted:** 26.10.2026 | **Published:** 02.11.2026

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Abstract

Plants are important source of nutrients and contain various chemical constituents that demonstrate nutritional, medicinal, and pharmacological properties. This study aimed at analyzing the chemical constituents present in the methanol seeds extract of *Persea americana*. The chemical constituents in methanol seeds extract of *Persea americana* were analyzed using Gas Chromatography-Mass Spectroscopy (GC-MS) technique. The GC-MS chromatogram of the extract showed a total of 20 putatively identified compounds. The chemical constituents identified in the extract with their percentage peak area were 5-Hydroxy-4-methyl-3-heptanone (22.82 %), decanoic acid (5.40 %), butoxyacetic acid (0.99 %), 6-methylheptyl vinyl (6.34 %), Docosyl octyl ether (1.73 %), 3-Deoxy-d-mannoic lactone (2.49 %), Oxazole (2.44 %), Hexadecanoic acid (2.90 %), 1,2-Benzenedicarboxylic acid (1.17 %), n-Hexadecanoic acid (12.10 %), 1-Octenylsuccinic anhydride (0.62 %), cis-13-Octadecenoic acid (5.40 %), 1H-Cycloprop[e]azulenedecahydro-1,1,4,7-tetramethyl- (6.18 %), cis-Vaccenic acid (12.62 %), Oleic acid (5.64 %), 7-Pentadecyne (2.33 %), 2-Propenoic acid (0.70 %), 6-Octadecenoic acid (2.60 %), 4-Thiazolidinone (1.54 %), and Glyceric acid (1.24 %). The methanol seeds extract of *Persea americana* contains several chemical compounds of biological importance.

Keywords: Bioactive compounds, GC-MS, *Persea americana*, Phytoconstituents.

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INTRODUCTION

Plants have been consumed as foods and high proportion of the world's population relies on plants and herbs for therapeutic of illness. More than 3.3 billion people in the world depend on plants and herbs for remedies ((Abubakar *et al.,* 2026; Wenji *et al.,* 2019). Plants and herbs contain several chemical constituents that responsible their pharmacological activities including antioxidant, anti-ulcer, and analgesic activities (Abubakar *et al.,* 2024; Abubakar *et al.,* 2021; Kumar *et al.,* 2021; Abubakar *et al.,* 2020a, Abubakar *et al.,* 2020b). Phytoconstituents are bioactive compounds that occur naturally in plants demonstrated different therapeutic properties (Zeeshan *et al.,* 2022; Bhuyan *et al.,* 2019). Plants chemical constituents have been used in drugs discovery and development (Aliyu *et al.,* 2025). Analysis of chemical constituents in plants has significant application in development of effective drugs for management of diseases.

Persea americana, commonly known as avocado is a perennial plant belonging to the family Lauraceae (Ejiofor *et al.,* 2018). Persea americana is a native to Central America and Mexico and is abundantly available in tropical and subtropical countries. Persea americana has received significant attention in both scientific research due to its high nutritional value, medicinal properties and economic importance. Persea americana seeds contains different phytoconstituents with different pharmacological activities suggesting its health benefits. Studies showed that bioactive compounds from Persea americana seeds exhibited significant antioxidant, anticancer, antiinflammatory, and antibacterial activities (Alkhalaf *et al.,* 2019; Dabas *et al.,* 2019). Persea americana seeds are important source of carbohydrates, fiber, protein, lipids, and vitamins (Féliz-Jiménez, 2024; Siol and Sadowska, 2023).

In Nigeria, *Persea americana* is locally known as Ube Oyinbo (Igbo), Piya (Hausa), and Ewe pia (Yoruba). The fruits of the plant have been commonly used in many communities in the country as food and for nutrition and medicine. Majority of individuals in the communities dispose the seeds of the plant post consumption of the fleshy part of the fruits. This habit

might be lack or insufficient information on its nutritional and medicinal benefits. Different parts of *Persea americana* have been used in management different diseases including malaria, fever, diabetes, ulcer, and cardiovascular disorders. Research indicated that chemical constituents of *Persea americana* significantly varied with environmental condition and techniques of cultivation or processing (Féliz-Jiménez and Sanchez-Rosario, 2024). This study aimed at analyzing the chemical constituents present in the methanol seeds extract of *Persea americana* cultivated in Owerri, Imo State, Nigeria.

MATERIALS AND METHODS

Chemicals and Reagents

All the chemicals and reagents used in this study were of analytical grades and manufactured by BDH Chemical Limited Poole (England, UK) and Sigma-Aldrich (St. Louis, MO, USA).

Plant Material

The *P. amercana* fruits were purchased from Ihiagwa Market, Owerri, Imo State, Nigeria. The samples were washed thoroughly with deionized water. The seeds were obtained by removal of fleshy part of the fruits. The seeds samples were dried overnight in an oven at 50 °C. The dried seeds sample were ground into powder using a mill and stored in a clean container.

Preparation of the Plant Extract

The methanol seeds extract of *Persea americana* was prepared according to the method of Abubakar *et al.* (2020a). The seeds powder sample (250 g) was soaked in 500 mL of methanol for 72 hours with constant stirring. The extract was filtered using Whatman filter paper No.1. The filtrate was concentrated in rotary evaporator under reduced pressure at 40 °C for 180 minutes. The weight and the percentage yield of the extract recorded were 13.42 g and 5.36 %, respectively.

GC-MS Analysis

The chemical constituents in the methanol seeds extract of *Persea americana* were analyzed using GC-MS equipment (Shimadzu Company's model GC/MS-QP2010-Ultra). The extract was treated with 5 mL of 96 % ethanol and ultrasonicated for at 55 ℃ for half hour. The extract was filtered and then injected into GC-MS operating (EI mode) capillary column (Rtx-5 ms-30m × 0.25mm × 0.25 µm) at 70 eV.A. Helium was used as the carrier gas at 1.69 mL/min flow rate. The temperature was initially set at 50 °C with a rate of 7 °C/min, and then 180 °C. The rate was increased to 10 °C/min, to a final temperature of 280 °C. The injection port temperature, ion source temperature, and the interface temperature was 300 °C, 200 °C, and 250 °C, respectively. The scan mode in the range of m/z 40–500 charges to ratio with a total run time of 28 minutes was used for the analysis.

Identification of Components

National Institute Standard Technology (NIST) database was used for the interpretation of the GC-MS mass spectrum. The database housed more than 62,000 patterns with library ID D:\LIBRARY\NIST17.L (Stein, 1990). The chemical constituents in the methanol seeds extract of *Persea americana* were identified by matching their retention time and mass spectra fragmentation patterns to those stored in the NIST. The name and the structure of the putative compounds in the extract were ascertained. The relative percentage composition of each putative compound was obtained from the relative peak area of each component in the chromatogram.

RESULT

GC-MS Analysis of Methanol Seeds Extract of *Persea americana*

Figure 1 shows the GC-MS chromatogram of methanol seeds extract of *Persea americana*. The chromatogram of the extract displayed twenty different peaks each indicating a specific chemical compound at a specific retention time.

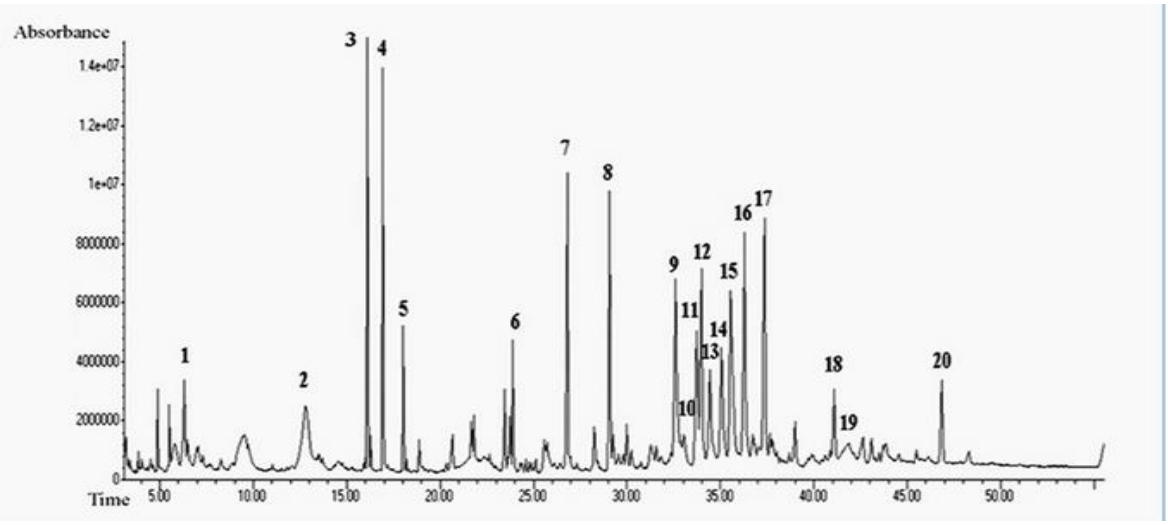
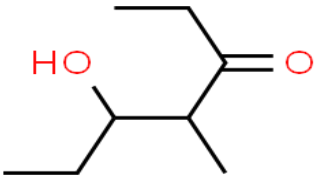
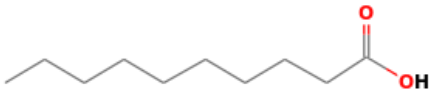
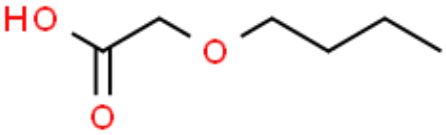
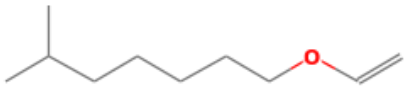
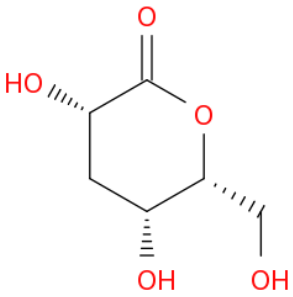


Figure 1: GC-MS Chromatogram of Methanol Seeds Extract of *Persea americana*

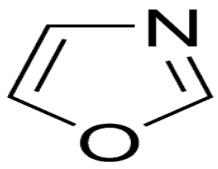
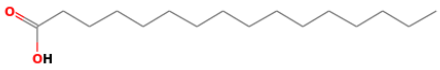
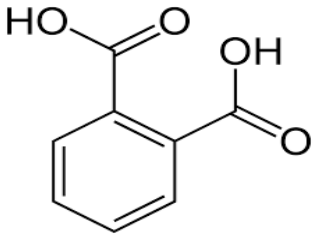
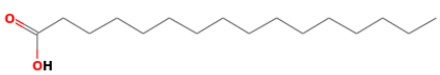
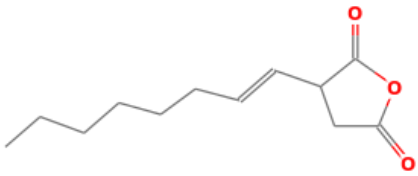
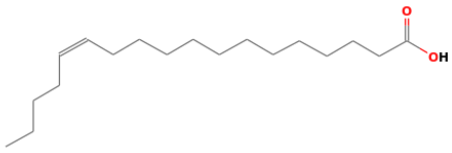
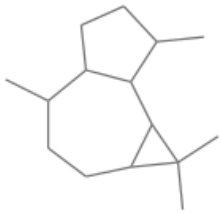
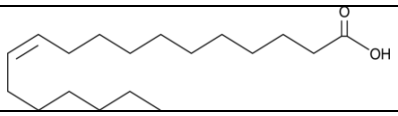
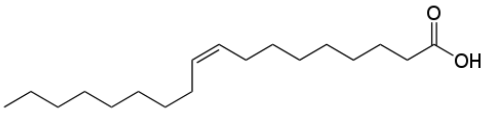
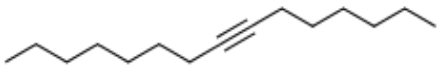
Putatively Identified Compounds in Methanol Seeds Extract of *Persea americana*

Table 1 shows the chemical constituents identified from the methanol seeds extract of *Persea americana* with their retention time, structures, and percentage peak area. A total of 20 chemical constituents were identified in the extract. The putatively identified compounds in the extract were 5-Hydroxy-4-methyl-3-heptanone (22.82 %), decanoic (capric acid) (5.40 %), butoxyacetic acid (sebacic acid) (0.99 %), 6-methylheptyl vinyl (6.34 %), Docosyl octyl ether (1.73 %), 3-Deoxy-d-mannoic lactone (2.49 %), Oxazole (2.44 %), Hexadecanoic acid (2.90 %), 1,2-Benzenedicarboxylic acid (1.17 %), n-Hexadecanoic acid (12.10 %), 1-Octenylsuccinic anhydride (0.62 %), cis-13-Octadecenoic acid (5.40 %), 1H-Cycloprop[e]azulenedecahydro-1,1,4,7-tetramethyl- (6.18 %), cis-Vaccenic acid (12.62 %), Oleic acid (5.64 %), 7-Pentadecyne (2.33 %), 2-Propenoic acid (0.70 %), 6-Octadecenoic acid (2.60 %), 4-Thiazolidinone (1.54 %), and Glyceric acid (1.24 %).

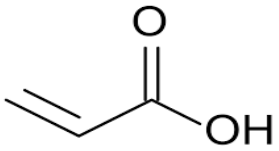
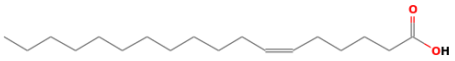
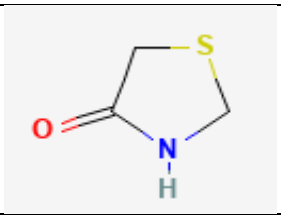
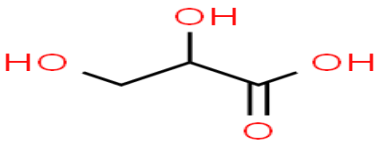
Table 1: Putatively Identified Compounds in Methanol Seeds Extract of *Persea americana*

S/No	Retention Time (min)	Compound	Structure	Relative Peak Area (%)
1	11.877	5-Hydroxy-4-methyl-3-heptanone		22.82
2	12.460	Decanoic acid		5.40
3	12.699	Butoxyacetic acid		0.99
4	14.435	6-methylheptyl vinyl		6.34
5	14.527	Docosyl octyl ether		1.73
6	14.703	3-Deoxy-d-mannoic lactone		2.49

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S/No	Retention Time (min)	Compound	Structure	Relative Peak Area (%)
7	15.474	Oxazole		2.44
8	16.982	Hexadecanoic acid		2.90
9	17.486	1,2-Benzenedicarboxylic acid		1.17
10	17.621	n-Hexadecanoic acid		12.10
11	18.208	1-Octenylsuccinic anhydride		0.62
12	18.800	cis-13-Octadecenoic acid		5.40
13	19.102	1H-Cycloprop[e]azulenedecahydro-1,1,4,7-tetramethyl-		6.18
14	19.425	cis-Vaccenic acid		12.62
15	19.604	Oleic Acid		5.64
16	20.268	7-Pentadecyne		2.33

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S/No	Retention Time (min)	Compound	Structure	Relative Peak Area (%)
17	20.564	2-Propenoic acid		0.70
18	23.602	6-Octadecenoic acid		2.60
19	23.931	4-Thiazolidinone		1.54
20	23.110	Glyceric acid		1.24

DISCUSSION

Plants and herbs have documented to demonstrate various phytoconstituents that are attributed to their nutritional and medicinal properties. In this study, 5-Hydroxy-4-methyl-3-heptanone, decanoic (capric acid), butoxyacetic acid (sebacic acid), 6-methylheptyl vinyl, Docosyl octyl ether, 3-Deoxy-d-mannoic lactone, Oxazole, Hexadecanoic acid, 1,2-Benzenedicarboxylic acid, n-Hexadecanoic acid, 1-Octenylsuccinic anhydride, cis-13-Octadecenoic acid, 1H-Cycloprop[e]azulenedecahydro-1,1,4,7-tetramethyl-, cis-Vaccenic acid, Oleic acid, 7-Pentadecyne, 2-Propenoic acid, 6-Octadecenoic acid, 4-Thiazolidinone, and Glyceric acid were detected in the methanol seeds extract of *Persea americana*. Decanoic acid or capric acid is a saturated fatty acid.

Capric acid (decanoic acid) oxidizes in hepatic mitochondria through β-oxidation to yield acetyl-CoA and ketone bodies that generate energy in the body. Capric acid acts as an AMPA receptor antagonist, PPAR-γ agonist, anti-inflammatory modulator, and antimicrobial agent (Sang *et al.,* 2017). Study indicated that capric acid significantly lowered the levels of TNF-α and IL-6 after cyclophosphamide treatment in porcine intestinal epithelial cells (Sang *et al.,* 2017). Capric acid inhibits inflammatory cytokines including IL-8, IL-6, and TNF-α by the inhibition of MAPK phosphorylation and NF-kB activation (Huang *et al.,* 2014). It has been documented that capric acid treatment alleviated oxidative stress induced by cyclophosphamide in small intestinal epithelial cells (Sang *et al.,* 2017).

Sebacic acid or butoxyacetic acid regulates inflammation by inhibiting NF-κB and MAPK pathways, improves glucose homeostasis and insulin sensitivity, participates in mitochondrial and peroxisomal β-oxidation, and promotes bone and muscle regeneration (Kunugi *et al.,* 2019). Sebacic acid modulates bone metabolism balance via the crosstalk between estrogen receptors and Wnt signaling pathways (Moutsatsou *et al.,* 2010). It is also used as a monomer for poly(glycerol sebacate) (PGS) to the development of biodegradable scaffolds for bone tissue engineering (Rai *et al.,* 2012). Study showed that sebacic acid demonstrates antidiabetic properties. Serum metabolomics analyses indicate that sebacic acid levels are significantly decreased in patients with diabetes and insulin resistance (Ling *et al.,* 2024). Endogenous sebacic acid is involved in fatty acid metabolism and energy production through the TCA cycle. Sebacic acid significantly decreased the production of tumor necrosis factor-α (TNF-α) (Chen *et al.,* 2016). Thus, sebacic acid remains an important agent for the management of chronic metabolic and inflammatory disorders and for applications in regenerative medicine.

Palmitic acid or hexadecanoic acid is the most common saturated fatty acid in the human, accounting for about 65 % of the saturated fatty acids and 28–32 % of the total fatty acids in serum (Carta *et al.,* 2017; Thomas and Huganir, 2013; Yu *et al.,* 2012). Palmitic acid demonstrated palmitoylation which serves as a switch that controls the protein's function, increases the hydrophobicity of membrane protein, and directs the protein to specific microdomains within the plasma

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membrane (Chamberlain *et al.,* 2013). Palmitoylation enhances transport of proteins between membrane organelles and within microdomains of individual membrane compartments, and, hence, modulates protein–protein interaction (Chamberlain *et al.,* 2013). Palmitic acid exhibits several pharmacological activities including antiviral, anti-inflammatory, analgesic, and regulation of lipid metabolism (Librán-Pérez *et al.,* 2019; Mayneris-Perxachs, 2014). Study showed that Palmitic acid has potential for inducing cell cycle arrest and enhancing apoptosis in human neuroblastoma and breast cancer cells (Hsiao *et al.,* 2014; Pereira *et al.,* 2013).

Petroselinic acid (Octadecenoic acid) is a rare fatty acid reported to displayed anti-inflammatory activity, inhibitory against topoisomerases, and for the modulatory effect of lipid metabolism (Hajib *et al.,* 2023). Investigation showed that petroselinic acid demonstrated preventive and therapeutic potential against intestinal ischemia-reperfusion injury (Li *et al.,* 2021). However, petroselinic acid improves the intestinal tissue injury induced by intestinal ischemia-reperfusion and the survival rate of the mice (Li *et al.,* 2021). Cis-vaccenic acid has been reported to reduce the production of pro-inflammatory cytokines, thus exhibiting anti-inflammatory properties (Matysik *et al.,* 2016). Cis-vaccenic acid serves important function in the management of diabetes (Tsalamandris *et al.,* 2019).

Oleic acids (Omega-9 fatty acids) exhibit various health benefits. Oleic acids serve therapeutic effect in management of diabetes mellitus. Oleic acids have been the most extensively studied for their roles in promoting human health (Neves *et al.,* 2024). Consumption of diet rich in oleic acids restored endothelium-dependent vasodilatation and decreased insulin resistance and type 2 diabetes mellitus (López-Gómez *et al.,* 2020; Palomer *et al.,* 2018). Similar study showed that Persea americana is highly rich in oleic acids (Lara-García *et al.,* 2021). A related research revealed that Persea americana contains high amount of oleic acids that demonstrated cardiovascular, anti-inflammatory, and cognitive benefits (García *et al.,* 2021). Oleic acids exhibited preventive properties against metabolic disturbances associated to alimentation of amino acids and hexose (Cury-Boaventura *et al.,* 2008; Waitzberg *et al.,* 2006). Research indicated that oleic acids displayed neuroprotective activities that may be due to its potential for activation of peroxisome proliferator-activated receptor gamma (PPAR-γ) (Katariya *et al.,* 2021; Song *et al.,* 2019).

GC-MS is an important technique used for analysis, identification, and quantification of compounds with high accuracy and reliability (Choudhury *et al.,* 2022). GC-MS is applicable for analyzing volatile and semi-volatile compounds. The technique is limited for the analysis of non-volatile or thermally unstable compounds (Beale *et al.,* 2018). The major limitation of GC-MS is the non-volatile nature of the primary metabolites (Zhang *et al.,* 2021; Liu *et al.,* 2020). Primary metabolites, such as carbohydrates, amino acids, and organic acids, are highly polar and cannot be efficiently retarded on capillary GC columns (Wang *et al.,* 2023; Zhang *et al.,* 2021; Liu *et al.,* 2020; Beale *et al.,* 2018). Due to this these metabolites cannot be analyzed with GC-MS directly without any modification of their structure prior to analysis.

CONCLUSION

The methanol seeds extract of *Persea americana* contains significant amount of 5-Hydroxy-4-methyl-3-heptanone, decanoic acid, butoxyacetic acid, 6-methylheptyl vinyl, Docosyl octyl ether, 3-Deoxy-d-mannoic lactone, Oxazole, Hexadecanoic acid, 1,2-Benzenedicarboxylic acid, n-Hexadecanoic acid, 1-Octenylsuccinic anhydride, cis-13-Octadecenoic acid, 1H-Cycloprop[e]azulenedecahydro-1,1,4,7-tetramethyl-, cis-Vaccenic acid, Oleic acid, 7-Pentadecyne, 2-Propenoic acid, 6-Octadecenoic acid, 4-Thiazolidinone, and Glyceric acid. These chemical constituents have been reported with different medicinal properties and pharmacological activities.

Conflict of Interest

The authors declared no conflict of interest.

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