

Phytochemical Composition and Therapeutic Potential of Ethanol Leaf Extract of *Annona muricata* from Nigeria Using Gas Chromatography–Mass Spectrometry

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Abstract- *Annona muricata* (Annonaceae) commonly known as soursop or graviola, is a medicinal plant extensively used in traditional medicine for the management of cancer, inflammation, infections, and metabolic disorders. This study investigated the phytochemical composition of *A. muricata* leaf extract using Gas Chromatography–Mass Spectrometry (GC–MS) and discussed the medicinal relevance of the identified compounds. GC–MS analysis revealed nineteen phytochemical constituents belonging to fatty acids, terpenoids, phenolics, tocopherols, alkynes, nitrogen-containing heterocycles, and long-chain hydrocarbon sterols, and hydrocarbons. Gamma-sitostenone and vitamin E were the predominant compounds with a percentage peak of (35.55% and 14.09%) respectively. Neophytadiene, 9-octadecenoic acid, 4-campestene-3-one, and n-hexadecanoic acid. Showed a percentage of (5.13%, 5.43%, 5.24% and 4.18%) respectively. Furthermore, the antioxidant, anti-inflammatory, antimicrobial, anticancer, and cardioprotective potential of *A. muricata* are evident in the Phytochemical found in its leaf. Evidence suggests that γ -sitostenone exhibit significant anti-inflammatory activity, while Vitamin E is widely known for its strong antioxidant activity. 9-Octadecenoic acid demonstrates cardioprotective potential, furthermore, neophytadiene and 4-campestene-3-one have been associated with notable antimicrobial and anticancer activities. Overall, these findings underscore the therapeutic relevance of *Annona muricata* leaf extract, particularly in relation to the diverse bioactive compounds responsible for its medicinal properties.

Keywords: *Annona Muricata*, GC–MS, Phytochemicals, Antioxidant, Medicinal Plants

I. INTRODUCTION

Medicinal plants remain an important source of bioactive compounds for drug discovery and complementary medicine, particularly in developing countries where traditional healthcare systems are widely practiced as reported in recent studies (Mutakin

et al., 2021; Adewole et al., 2022; Coria-Téllez et al., 2022). *Annona muricata*, commonly known as soursop or graviola, is widely distributed in tropical regions and has gained scientific interest due to its diverse pharmacological properties. Traditionally, the leaves of *A. muricata* are used to treat fever, pain, infections, diabetes, hypertension, and cancer (Adewole et al., 2022).

Previous studies have attributed these biological activities to the presence of secondary metabolites such as acetogenins, alkaloids, flavonoids, terpenoids, fatty acids, sterols, and phenolic compounds (Mutakin et al., 2021; Coria-Téllez et al., 2022). While acetogenins are well studied, volatile and semi-volatile constituents identifiable by Gas Chromatography–Mass Spectrometry (GC–MS) also play critical roles in the medicinal activity of the plant (Olatunji et al., 2023).

GC–MS is a powerful analytical technique for rapid identification of phytochemicals and provides insight into the chemical diversity responsible for biological effects (Olatunji et al., 2023). This study aimed to profile the phytochemical Present in *A. muricata* Ethanol leaf extract and its medicinal relevance.



Figure 1. refers to the various plant parts of *Annona muricata*: (A) whole plant, (B) leaves, (C) flower, (D) fruit, (E) seeds. Source: (Coria-Téllez *et al.*, 2022).

II. MATERIALS AND METHODS

2.1 Plant Material and Extraction

Fresh leaves of *Annona muricata* were collected, washed, air-dried at room temperature, and pulverized into powder. The powdered material was extracted using an organic solvent (ethanol) by maceration. The extract was filtered, concentrated, and stored at 4 °C prior to analysis.

2.2 GC–MS Analysis

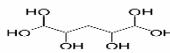
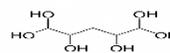
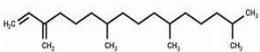
GC–MS analysis was carried out using a capillary column coupled to a mass spectrometer operating under electron ionization mode. Helium was used as the carrier gas, and the oven temperature was programmed gradually to allow separation of compounds. Phytochemicals were identified by comparing their mass spectra with those in the NIST library, alongside retention time and molecular weight confirmation, following standard analytical procedures (Olatunji *et al.*, 2023).

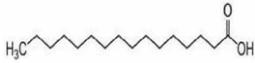
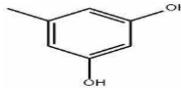
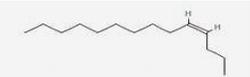
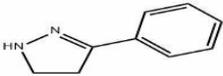
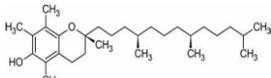
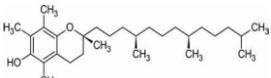
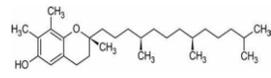
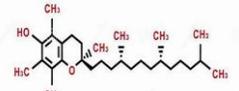
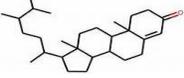
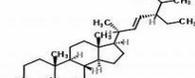
III. RESULTS

GC–MS analysis of *A. muricata* leaf extract revealed nineteen (19) phytochemical constituents with varying retention times and relative peak areas. The identified compounds belonged to multiple chemical classes including polyols, terpenoids, fatty acids, phenolics, tocopherols, and sterols.

Gamma-sitostenone (35.55%) was the most abundant compound, followed by vitamin E (14.09%). Other notable constituents included neophytadiene (5.13%), 9-octadecenoic acid (5.43%), 4-campestene-3-one (5.24%), n-hexadecanoic acid (4.18%), and stigmasteroid derivatives.

3.1 Table of Phytochemical composition of *A. muricata* ethanol leaf extract

S/N	Name Of Compound	Retention Time (Rt)	Molecular Formular(Mf)	Molecular Weight (Mw)	Percentage (%)	Structure
1	1,2,3,4-Butanetetrol	3.056	C ₄ H ₁₀ O ₄	122.12 g/mol	2.64	
2	1,2,3,4-Butanetetro	4.853	C ₄ H ₁₀ O ₄	122.12 g/mol	1.85	
3	Neophytadiene	5.877	C ₂₀ H ₃₈	278.52 g/mol	5.13	
4	3-Eicosyne	5.997	C ₂₀ H ₃₈	278.52 g/mol	1.84	
5	Cyclohexene, nitro-	1- 6.089	C ₆ H ₉ NO ₂	127.87 g/mol	6.77	

6	n-Hexadecanoic acid	6.735	C ₁₆ H ₃₂ O ₂	256.23 g/mol	4.18	
7	Orcinol	6.959	C ₇ H ₈ O ₂	124.97 g/mol	2.33	
8	4-Tetradecyne	7.136	C ₁₄ H ₂₆	194.51 g/mol	1.28	
9	9-Octadecenoic acid	7.433	C ₁₈ H ₃₄ O ₂	282.88 g/mol	5.43	
10	1H-Pyrazole, 4,5-dihydro-3-phenyl-	8.069	C ₉ H ₁₀ N ₂	146.98 g/mol	2.34	
11	Alpha.-Tocospiro B	9.854	C ₂₉ H ₅₀ O ₄	462.91 g/mol	1.31	
12	Alpha.-Tocospiro B	9.934	C ₂₉ H ₅₀ O ₄	462.91 g/mol	1.51	
13	Gamma.-Tocopherol	11.324	C ₂₈ H ₄₈ O ₂	416.43 g/mol	2.11	
14	Vitamin E	11.857	C ₂₉ H ₅₀ O ₂	430.08 g/mol	14.09	
15	Phenyl p-tolyethynyl ketone	14.466	C ₁₆ H ₁₂ O	220.15 g/mol	1.61	
16	benzenesulfonamide, 3-amino-N-methyl-N-octadecyl-	14.952	C ₂₅ H ₄₆ N ₂ O ₂ S	438.28 g/mol	1.30	
17	4-Campestene-3-one	15.507	C ₂₈ H ₄₆ O	398.66 g/mol	5.24	
18	4,22-Stigmastadiene-3-one	15.679	C ₂₉ H ₄₆ O	410.66 g/mol	3.49	
19	Gamma.-Sitostenone	16.360	C ₂₉ H ₄₂ O	412.16 g/mol	35.55	

IV. DISCUSSION

The phytochemical composition of *Annona muricata* ethanol leaf extract reveals a diverse array of bioactive compounds that collectively contribute to its antioxidant, antimicrobial, anti-inflammatory,

cardioprotective, and anticancer properties. 1,2,3,4-Butanetetrol is a polyhydric alcohol exhibits antioxidant and free-radical scavenging properties. Recent studies highlight its role in protecting cells against oxidative stress and supporting metabolic balance (Olatunji, Afolayan, & Bradshaw, 2022)

(Olatunji, Afolayan, & Bradshaw, 2022). However, the isomer with structural similarities contributes to redox homeostasis and may enhance cellular defense mechanisms against oxidative damage. Neophytadiene is a diterpenoid hydrocarbon reported to possess anti-inflammatory, antimicrobial, and anticancer activities. Recent evidence indicates its ability to inhibit inflammatory mediators and microbial growth (Nolasco-González et al., 2022; Mendes, Silva, & Fernandes, 2024) (Nolasco-González et al., 2022; Mendes, Silva, & Fernandes, 2024). Furthermore, 3-Eicosyne is a Long-chain alkynes that have demonstrated antimicrobial and insecticidal properties. They may contribute to the traditional use of *A. muricata* leaves in managing infections (Zahid et al., 2022) (Zahid et al., 2022). Cyclohexene, 1-nitro, a nitro-substituted cyclohexene, has been associated with antimicrobial and cytotoxic activities, indicating potential relevance in anticancer drug development.

Fatty acids identified in the extract further reinforce its therapeutic value. n-Hexadecanoic acid (palmitic acid) exhibits antimicrobial, antioxidant, and anti-inflammatory properties and plays a role in modulating immune responses and inhibiting pathogenic microorganisms. Likewise, 9-Octadecenoic acid (oleic acid), a monounsaturated fatty acid, demonstrates cardioprotective, anti-inflammatory, and anticancer effects, with recent studies highlighting its capacity to regulate lipid metabolism and reduce oxidative stress (Chukwuma, Matsabisa, & Ibrahim, 2024).

Phenolic and aromatic compounds also contribute significantly to the biological activity of the extract. Orcinol, a phenolic compound, shows strong antioxidant, antimicrobial, and anti-inflammatory properties, with emerging evidence suggesting neuroprotective and anticancer potential through modulation of oxidative stress pathways (Eze et al., 2022; Khan, Ahmad, & Asiri, 2023). Phenyl p-tolyl vinyl ketone has demonstrated antimicrobial and cytotoxic activities, supporting possible anticancer applications (Olatunji et al., 2022; Khan et al., 2023). Benzenesulfonamide, 3-amino-N-methyl-N-octadecyl-, a sulfonamide derivative, is recognized for broad-spectrum antimicrobial, anti-inflammatory, and

anticancer effects, largely attributed to its enzyme-inhibitory potential.

The presence of alkynes such as 4-Tetradecyne, known for antimicrobial and anti-biofilm properties, further supports the plant's traditional use against infections. In addition, heterocyclic compounds like 1H-Pyrazole, 4,5-dihydro-3-phenyl- exhibit significant pharmacological activities, including anticancer, anti-inflammatory, and antimicrobial effects, and are widely recognized as important scaffolds in modern drug discovery (Khan et al., 2023).

A prominent feature of the extract is the abundance of tocopherol-related compounds and phytosterols. Alpha-Tocospiro B and its isomer display antioxidant and lipid-protective properties that enhance cellular membrane stability and prevent oxidative damage (Al-Snafi, 2022; Mendes et al., 2024). Gamma-tocopherol, a potent form of vitamin E, demonstrates strong antioxidant, anti-inflammatory, cardioprotective, and cancer-preventive effects, alongside immunomodulating functions (Al-Snafi, 2022; Mendes et al., 2024). Vitamin E itself is widely recognized for its neuroprotective, cardioprotective, anti-aging, and lipid-peroxidation-inhibiting properties (Al-Snafi, 2022).

Phytosterol derivatives such as 4-Campestene-3-one and 4,22-Stigmastadiene-3-one contribute anti-inflammatory, hypolipidemic, antioxidant, antidiabetic, and anticancer activities, thereby supporting metabolic and cardiovascular health (Mendes et al., 2024). Gamma-Sitostenone further exhibits strong anti-inflammatory, anticancer, and hypocholesterolemic effects, reinforcing the therapeutic relevance of *A. muricata* leaves (Adewole et al., 2023; Uchegbu, Okafor, & Ibeanu, 2025).

4.1 Synergistic Pharmacological Effects

The coexistence of fatty acids, terpenoids, phenolics, tocopherols, and sterols suggest synergistic interactions that enhance the overall medicinal activity of the extract. Such synergy is common in medicinal plants and supports their multi-target therapeutic potential (Munteanu & Apetrei, 2021; Adewole et al., 2022).

V. MEDICINAL IMPORTANCE OF ANNONA MURICATA LEAF EXTRACT

Based on the identified phytochemicals, *A. muricata* leaf extract exhibits:

Antioxidant activity: due to tocopherols, phenolics, and unsaturated fatty acids (Munteanu & Apetrei, 2021; Jiang, 2024).

Anticancer potential: supported by sterols, terpenoids, and antioxidant compounds (Mutakin et al., 2021)

Anti-inflammatory effects: through modulation of inflammatory mediators (Adewole et al., 2022)

Antimicrobial properties: via membrane-disrupting fatty acids and terpenoids (Olatunji et al., 2023)

Cardioprotective and metabolic benefits: associated with phytosterols and oleic acid Coria-Téllez et al., 2022)

VI. CONCLUSION

GC–MS profiling of *Annona muricata* leaf extract revealed a diverse range of bioactive phytochemicals, with gamma-sitostenone and vitamin E as dominant constituents. The identified compounds possess well-documented antioxidant, anti-inflammatory, antimicrobial, anticancer, and cardioprotective properties. These findings provide scientific validation for the traditional medicinal use of *A. muricata* leaves and highlight their potential as a source of bioactive compounds for pharmaceutical development.

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