

## Phytochemical Profiling of *Opa eyin*, a Traditional Nigerian Herbal Preparation, by GC–MS and Its Potential Pharmacological Implications

Uduak I. Aletan\* and Sunday Adenekan

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**Abstract:** *This study investigated the phytochemical composition of Opa eyin, a traditional herbal remedy commonly used in southwestern Nigeria for the management of erectile dysfunction and back pain. Gas chromatography–mass spectrometry (GC–MS) analysis revealed 115 distinct peaks corresponding to a diverse array of bioactive compounds. Among these, twenty major phytoconstituents were identified, with methyl gallate [benzoic acid, 3,4,5-trihydroxy-, methyl ester] as the predominant component at relative abundances of 17.93% and 11.50%. Other abundant compounds included 11-octadecenoic acid methyl ester (10.18%), 1,2,3-benzenetriol (7.71%), and azelaic acid (6.57%), while moderate levels of methyl stearate (2.45%), oleic acid (2.53%), hexadecanoic acid methyl ester (2.25%), and terephthalic acid (1.77%) were also detected. The phytochemical profile indicates enrichment in phenolic and fatty acid derivatives, classes of compounds recognized for their antioxidant, anti-inflammatory, and cytoprotective properties. These findings suggest that the ethnomedicinal use of Opa eyin may be attributed to the synergistic actions of its phenolic antioxidants and fatty acids, which could support endothelial function, reduce oxidative stress, and alleviate inflammation. The results provide preliminary scientific evidence supporting its traditional applications; however, further in vivo studies and clinical evaluations are recommended to validate its efficacy and establish its safety profile..*

**Keywords:** *Opa eyin, phytochemical profiling, GC–MS, erectile dysfunction, ethnopharmacology*

Uduak I. Aletan\*

Department of Biological Sciences, Faculty of Sciences, National Open University of Nigeria, Jabi, Abuja, Nigeria

Email: [ualetan@noun.edu.ng](mailto:ualetan@noun.edu.ng)

ORCID ID : <https://orcid.org/0000-0003-0071-9632>

Sunday Adenekan

Department of Biochemistry, University of Lagos, Lagos, Nigeria

Email: [sundayadenekan2@gmail.com](mailto:sundayadenekan2@gmail.com)

ORCID ID : <https://orcid.org/0009-0000-9132-8471>

### 1.0 Introduction

Traditional herbal medicine has constituted a fundamental component of human healthcare for centuries and remains relevant in contemporary medical practice (WHO, 2023). This role is particularly pronounced in developing countries, where limited access to conventional health services and essential medicines often necessitates alternative therapeutic options. A considerable proportion of the population continues to depend on herbal remedies, largely due to their affordability (Tumuhaise et al., 2021) and the widespread perception of minimal toxicity (Van Wyka & Prinsloo, 2020). *Opa eyin*, a term in Yoruba that translates literally as ‘back rod,’ denotes a traditional herbal formulation commonly employed for the management of back pain and erectile dysfunction. Within urban centres of southwestern Nigeria, it is frequently marketed by young women as part of the broader informal trade in indigenous medicinal preparations.

Previous investigations have provided scientific insights into the safety profile of *Opa eyin*. Aletan et al. (2022) conducted a sub-chronic toxicity study on the formulation,

while subsequent work by Aletan et al. (2025) evaluated its in vitro and antioxidant-related toxicological properties. Despite these research efforts and the widespread use of *Opa eyin*, the scientific literature remains limited regarding its specific phytochemical composition.

Comprehensive characterization of its chemical profile is essential, not only to substantiate its ethnomedicinal applications and establish safety but also to facilitate the identification of novel bioactive compounds with potential pharmaceutical relevance.

The analysis of complex herbal mixtures constitutes a critical step in phytochemical research and is commonly facilitated by advanced analytical platforms. Among these, Gas Chromatography–Mass Spectrometry (GC–MS) has emerged as a particularly robust technique, enabling the separation of volatile and semi-volatile constituents and their subsequent identification through characteristic mass spectral fragmentation patterns (Fiehn, 2016). Therefore, applying GC–MS to *Opa eyin* offers a valuable opportunity to generate a detailed phytochemical profile, which may serve as a foundation for correlating its traditional uses with scientifically validated bioactive components.

Similar phytochemical investigations using GC–MS have demonstrated the therapeutic potential of other African and Asian medicinal plants. For instance, Idam et al. (2022) profiled *Uvaria ovata* root extract and identified alkaloids, tannins, flavonoids, and eleven key compounds such as D-streptamine and cystine with significant medicinal potential. Ahamefula, Ordu, and Oruamabo (2020) reported antioxidant activities linked to phytochemicals in *Gongronema latifolium*, *Petrocarpus mildbraedii*, and *Piper guineense*, while Shemishere et al. (2025) characterized 28 bioactive compounds in Jinja™ herbal extract that demonstrated strong antioxidant properties. Beyond African contexts, Khan et al. (2020) identified fifty-seven phytoconstituents in *Chukrasia velutina* leaves, several of which

showed promising neuropsychiatric effects through in vivo rodent models and in silico docking analyses. These studies highlight the relevance of GC–MS in linking phytochemical profiles to pharmacological activities, reinforcing its importance in validating the therapeutic claims of traditional remedies.

This study, therefore, aims to investigate the phytochemical composition of *Opa eyin* using GC–MS, with the goal of identifying its major bioactive constituents and providing scientific evidence that supports its traditional medicinal applications. By situating *Opa eyin* within the broader context of phytochemical studies on medicinal plants, this work also underscores the significance of characterizing indigenous formulations as potential sources of antioxidant, anti-inflammatory, and reproductive health-enhancing agents.

## 2.0 Material and Methods

### 2.1 Sample collection

The sample of *Opa eyin* was obtained from a traditional medicine hawker in the Mushin market in the Mushin Local Government Area of Lagos State, Nigeria. There was no evidence of official registration of this remedy. The hawkers interviewed were not willing to give out their recipes due to their oath of secrecy. The purchased sample was stored in sterile amber glass containers at 4 °C until analysis to prevent degradation of volatile compounds.

### 2.2 Sample Preparation

Prior to GC–MS analysis, 5 mL of the liquid preparation was filtered through Whatman No. 1 filter paper to remove insoluble materials. The filtrate was then subjected to solvent extraction using analytical grade methanol (1:5, v/v), vortex-mixed for 5 min, and centrifuged at 4000 rpm for 10 min. The supernatant was concentrated under reduced pressure at 40 °C and reconstituted in 1 mL of methanol for injection. All analyses were performed in triplicate to ensure reproducibility.

### 2.3 Phytochemical Profiling Using GC–MS



The GC–MS analysis was performed on an Agilent 7820A gas chromatograph coupled to a 5975C inert mass spectrometer with a triple-axis detector and electron impact (EI) ion source (Agilent Technologies). Instrument tuning was conducted with perfluorotributylamine (PFTBA) to confirm ion abundances at  $m/z$  69, 219, and 502. Separation was achieved on an HP-5 capillary column (30 m  $\times$  0.32 mm  $\times$  0.25  $\mu$ m, 5% phenyl methyl siloxane; Agilent Technologies) using helium as the carrier gas at a constant flow of 1.49 mL/min.

Samples (1  $\mu$ L) were injected in splitless mode at 300  $^{\circ}$ C. The oven was programmed from 40  $^{\circ}$ C (1 min), ramped at 12  $^{\circ}$ C/min to 300  $^{\circ}$ C, and held for 10 min (total run time 32.7 min; solvent delay 5 min). The MS was operated in EI mode (70 eV), with ion source, quadrupole, and transfer line temperatures of 230, 150, and 280  $^{\circ}$ C, respectively. Data were acquired in scan mode over  $m/z$  45–550.

Compound identification was carried out by matching mass spectra with the National Institute of Standards and Technology (NIST 14) mass spectral library, with only matches above 80% similarity index considered valid. Where possible, retention indices were compared with published literature to strengthen identification confidence. Relative abundance of compounds was expressed as a percentage of the total ion chromatogram peak area.

A procedural blank (methanol) was run under identical conditions to eliminate background

interference, and results were corrected accordingly.

### 3.0 Results and Discussion

Gas chromatography–mass spectrometry (GC–MS) analysis of *Opa eyin* revealed 115 distinct peaks, representing a diverse array of phytochemicals. Of these phytochemicals, 20 major phytoconstituents have been presented on Table 1. The predominant constituents were benzoic acid, 3,4,5-trihydroxy-, methyl ester (methyl gallate) (17.93%, 11.50%), 11-octadecenoic acid methyl ester (10.18%), 1,2,3-benzenetriol (7.71%), and azelaic acid (6.57%). Other notable compounds included methyl stearate (2.45%), oleic acid (2.53%), octadecanoic acid (0.62%), as well as phenolic derivatives such as catechol and 1,2,4-benzenetriol. In addition, appreciable levels of fatty acid esters (e.g., hexadecanoic acid methyl ester, 2.25%) and aromatic acids (terephthalic acid derivatives) were detected. Overall, the phytochemical profile indicates that the concoction is particularly enriched in phenolic and fatty acid derivatives, classes of compounds well recognized for their antioxidant, antimicrobial, and anti-inflammatory activities. The table summarizes the identified compounds, their retention times, and their relative abundance (Area%). This list includes the most abundant compounds along with other notable identifications.

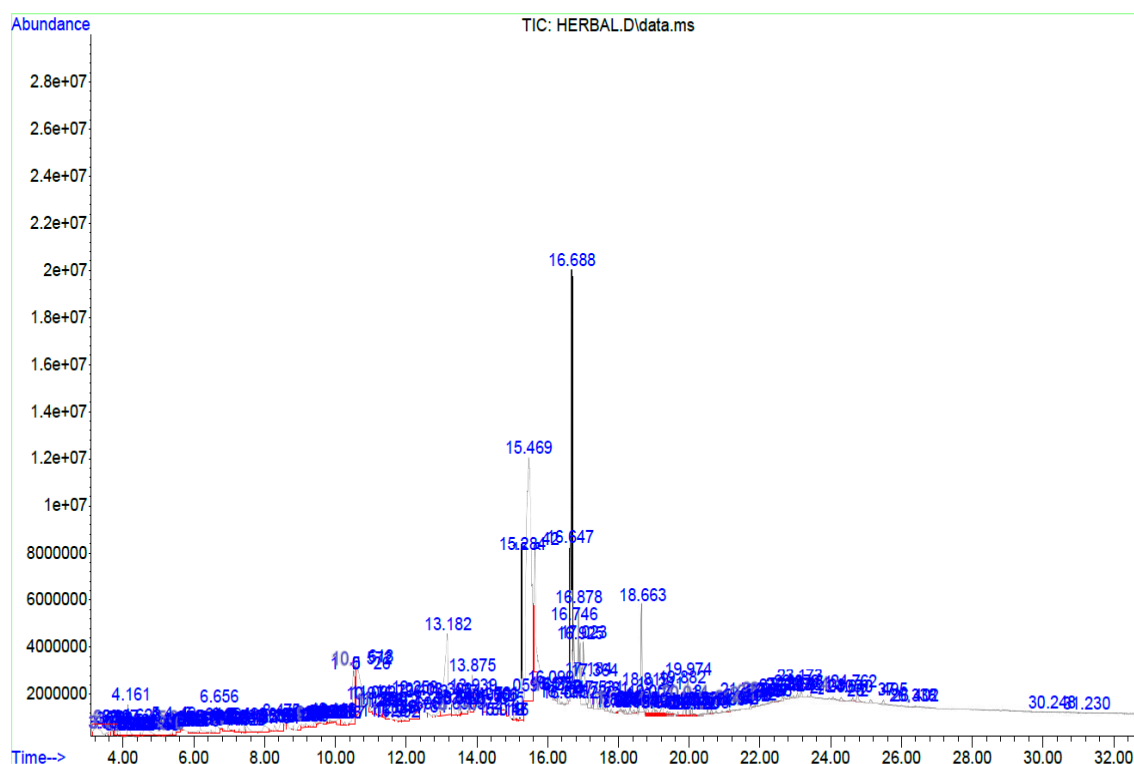
**Table 1. Major phytochemical constituents of *Opa eyin* identified by GC–MS analysis**

Peak #	Retention Time (RT)	Compound Name	Area%
30	10.618	1,2,3-Benzenetriol	7.71
40	13.182	Azelaic Acid	6.57
49	15.284	Hexadecanoic acid, methyl ester	2.25
50	15.469	Benzoic acid, 3,4,5-trihydroxy-, methyl ester	17.93
51	15.642	Benzoic acid, 3,4,5-trihydroxy-, methyl ester	11.5
55	16.688	11-Octadecenoic acid, methyl ester	10.18
57	16.878	Methyl Stearate	2.45
58	17.023	9-Octadecenoic acid, (E)-	2.53
69	18.663	9-Octadecenamide, (Z)-	1.86



11	6.194	Acetamide, N-[4-(trimethylsilyl)ph]enyl]-	0.26
22	8.475	Catechol	0.84
28	9.856	.beta.-D-Ribopyranoside, methyl	0.47
29	10.116	.alpha.-D-Galactopyranoside, methyl	0.63
37	12.258	Octanedioic acid	2.38
38	12.403	Nonanedioic acid, monomethyl ester	1.91
44	13.875	Benzene, 1,1'-(1,2-cyclobutanediyl)bis-, trans-	3.06
47	14.701	Terephthalic acid	1.77
60	17.364	9-Octadecenamide, (Z)-	0.84
102	24.762	Ethanone, 2-(2-benzothiazolylthio)-1-(3,5-dimethylpyrazolyl)-	0.89
104	25.495	2-Ethylacridine	0.27

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**Fig 1. Total ion chromatogram (TIC) of *Opa eyin* obtained by GC–MS**

The major peaks correspond to benzoic acid, 3,4,5-trihydroxy-, methyl ester (RT 15.47 and 15.64 min), 11-octadecenoic acid methyl ester (RT 16.69 min), 1,2,3-benzenetriol (RT 10.62 min), and azelaic acid (RT 13.18 min), which represent the predominant constituents of the extract.

Gas chromatography–mass spectrometry (GC–MS) analysis of *Opa eyin*, a traditional

herbal medicine commonly used in southwestern Nigeria for the management of erectile dysfunction and back pain, revealed 115 peaks corresponding to diverse classes of phytochemicals. The predominant compounds included benzoic acid, 3,4,5-trihydroxy-, methyl ester (methyl gallate) (17.93%, 11.5%), 11-octadecenoic acid methyl ester (10.18%), 1,2,3-benzenetriol (pyrogallol) (7.71%), and azelaic acid





(6.57%). Additional constituents such as methyl stearate, oleic acid, octadecanoic acid, and terephthalic acid were detected in moderate amounts. These findings are consistent with the rich phenolic and fatty acid composition of medicinal plants traditionally associated with antioxidant and anti-inflammatory properties (Sun & Shahrajabian, 2023)

Phenolic constituents such as methyl gallate and pyrogallol are well-established antioxidants capable of scavenging reactive oxygen species (ROS) and modulating inflammatory signaling pathways (Merecz-Sadowska et al., 2021). Methyl gallate in particular has been reported to exert strong free radical-scavenging, anti-inflammatory, and cytoprotective effects (Liang et al., 2023). The presence of these compounds in *Opa eyin* provides mechanistic support for its traditional use in improving male reproductive health, as oxidative stress and inflammation are central to the pathophysiology of erectile dysfunction (Kaltsas et al., 2024). By preserving nitric oxide (NO) bioavailability and endothelial function, phenolic antioxidants may enhance penile vascular relaxation and erectile capacity.

This interpretation aligns with findings from our previous work, where *Opa eyin* demonstrated significant *in vitro* antioxidant capacity and improved reproductive indices in *in vivo* models (Aletan et al., 2025). In that study, among other things the testicular antioxidant status, corroborating the present GC–MS evidence that phenolic phytoconstituents are key contributors to its reproductive health benefits.

In addition to polyphenols, the GC–MS profile revealed the presence of fatty acids and esters, such as hexadecanoic acid methyl ester, methyl stearate, and oleic acid derivatives. Fatty acids have been associated with anti-inflammatory and membrane-stabilizing activities, while certain unsaturated fatty acids like oleic acid protect endothelial cells against oxidative stress (Mallick & Duttaroy, 2022) These

compounds may contribute to the analgesic and anti-inflammatory actions traditionally attributed to *Opa eyin*, supporting its use in the management of back pain. Collectively, these findings suggest that the pharmacological actions of *Opa eyin* can be attributed to synergistic effects of phenolic antioxidants, fatty acids, and dicarboxylic acids. The antioxidant and anti-inflammatory mechanisms of these constituents provide a plausible scientific rationale for the ethnomedicinal use of the concoction in erectile dysfunction and back pain. Importantly, the integration of phytochemical profiling with prior *in vitro* and *in vivo* studies (Aletan et al., 2023) strengthens the evidence base for its therapeutic potential.

## 5.0 Conclusion

The study revealed that *Opa eyin*, a traditional herbal remedy, contains a rich phytochemical profile with 115 compounds identified through GC–MS analysis, of which methyl gallate, 11-octadecenoic acid methyl ester, pyrogallol, and azelaic acid were the predominant constituents. These phytochemicals belong mainly to phenolic and fatty acid derivatives, which are widely known for their antioxidant, anti-inflammatory, and cytoprotective properties. The findings suggest that the traditional use of *Opa eyin* for erectile dysfunction and back pain may be scientifically justified, as the identified compounds provide mechanistic support for improving reproductive health and alleviating inflammation-related conditions. Based on these results, it can be concluded that the pharmacological potential of *Opa eyin* lies in the synergistic actions of its bioactive constituents, particularly phenolic antioxidants and fatty acids. However, while the GC–MS analysis provides useful insights into its phytochemical composition, further experimental validation, including bioassays and clinical studies, is necessary to confirm its efficacy and safety. It is therefore recommended that comprehensive toxicological studies, *in vivo* investigations, and controlled clinical trials be conducted to



establish dosage, safety margins, and therapeutic applications of *Opa eyin* for its integration into evidence-based complementary and alternative medicine.

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### **Consent for publication**

Not applicable

### **Availability of data**

Data shall be made available on demand.

### **Competing interests**

The authors declared no conflict of interest

### **Ethical Consideration**

Not applicable

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### **Authors Contribution**

UIA conceived the work , literature search and prepared the manuscript, SA managed the analysis and data interpretation, Both authors read through the final manuscript.

