Research Article **Open Access**

Phytochemical screening and antioxidant evaluation of medicinal plants used for the menstrual pain management in Dekina local government area of Kogi state, Nigeria

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Editorial Office: Editor@FFHDJ.com

Submission Date: January 29th, 2026; **Acceptance Date:** March 24th, 2026; **Publication Date:** April 1st, 2026

Please cite this article as: Ocholi H. A., Ojochide A. D., Boniface M. T., David Z. A. Phytochemical screening and antioxidant evaluation of medicinal plants used for the menstrual pain management in Dekina local government area of Kogi State, Nigeria. *Agriculture and Food Bioactive Compounds.* 2026; 3(4): 54-64. DOI: https://doi.org/10.31989/afbc.v3i4.1923

ABSTRACT

Background: Dysmenorrhea, affecting 50–90% of reproductive-aged women globally, remains inadequately managed through conventional pharmacotherapy due to associated adverse effects.

Objective: This study aimed to document medicinal plants used traditionally for menstrual pain management in Dekina LGA, Kogi State, and to chemically profile and evaluate their antioxidant potential.

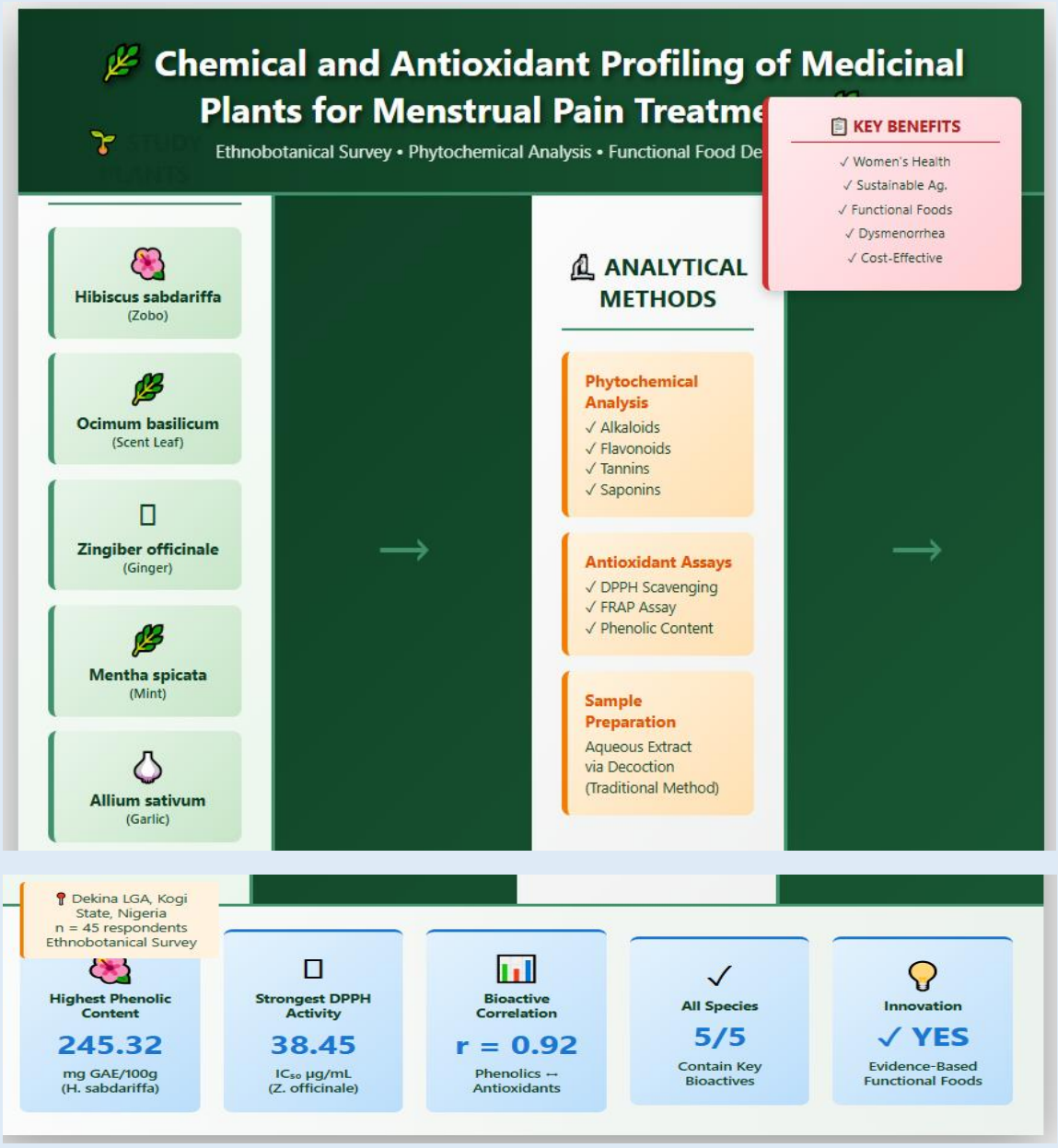
Methods: Ethnobotanical surveys were conducted with 45 traditional healers and herbalists (March–August 2023). Twenty-three plant species from 19 families were documented. Five frequently cited species (≥60% respondent consensus) were selected for analysis: *Hibiscus sabdariffa* L., *Ocimum basilicum* L., *Zingiber officinale* Roscoe, *Mentha spicata* L., and *Allium sativum* L. Aqueous extracts were prepared by decoction and analyzed for phytochemical constituents using standard colorimetric methods. Antioxidant activities were evaluated using DPPH radical scavenging, FRAP assay, and total phenolic content determination.

Results: Results showed that *H. sabdariffa* exhibited the highest total phenolic content (245.32±8.21 mg GAE/100g dry weight), while *Z. officinale* demonstrated the strongest DPPH radical scavenging activity (IC_{50} = 38.45±2.15 µg/mL). All species contained significant levels of alkaloids, flavonoids, tannins, and saponins.

Conclusion: These findings provide scientific validation for the traditional use of these plants and suggest their potential as complementary therapeutics for dysmenorrhea management

Novelty of Study: This study represents the first systematic ethnobotanical documentation of medicinal plants used for dysmenorrhea management in Dekina Local Government Area, Kogi State, Nigeria

Keywords: dysmenorrhea, phytochemical profiling, antioxidant activity, ethnobotany, bioactive compounds, medicinal plants



Graphical Abstract: Phytochemical screening and antioxidant evaluation of medicinal plants used for the menstrual pain management in Dekina local government area of Kogi state, Nigeria.

INTRODUCTION

Dysmenorrhea, commonly referred to as menstrual pain or painful menstruation, is characterized by recurrent cramping abdominal and pelvic pain during or immediately preceding menstruation. This condition affects approximately 50–90% of women of reproductive age globally, with 10–15% experiencing severe symptoms that significantly impair daily activities and work productivity [1]. Primary dysmenorrhea, occurring without identifiable pelvic pathology, is predominantly associated with elevated uterine prostaglandin levels and increased myometrial contractility, while secondary dysmenorrhea results from underlying reproductive disorders such as endometriosis and adenomyosis [2].

Conventional pharmaceutical management involves nonsteroidal anti-inflammatory drugs (NSAIDs) and hormonal contraceptives; however, these treatments are associated with significant adverse effects, including gastrointestinal complications, cardiovascular risks, and variable treatment efficacy [3]. Consequently, there is growing clinical and research interest in alternative and complementary therapeutic approaches, particularly plant-based remedies that demonstrate efficacy with minimal reported side effects [4].

In Sub-Saharan Africa, medicinal plants are a primary source of healthcare, with over 80% of the population relying on traditional medicine for treating various ailments, including menstrual disorders [5]. Nigeria, with its rich biodiversity and ethnobotanical heritage, contains numerous plant species with potential therapeutic applications. Recent comprehensive characterization studies of traditional foods and medicinal plants from Nigeria have demonstrated the substantial bioactive compound content and functional properties of indigenous species [16,18-19], yet scientific documentation and chemical validation of traditionally used medicinal plants for dysmenorrhea treatment remain limited, particularly in underexplored regions such as Dekina LGA in Kogi State.

Recent evidence demonstrates that oxidative stress plays a significant pathophysiological role in dysmenorrhea, with elevated reactive oxygen species (ROS) production and reduced antioxidant enzyme activity observed in women with severe pain symptoms [6]. Medicinal plants rich in polyphenolic compounds and antioxidants may provide therapeutic benefits through multiple mechanisms, including inhibition of prostaglandin synthesis, ROS scavenging, and modulation of inflammatory pathways [7]. Recent studies on green synthesis of bioactive agents have demonstrated the efficacy of natural compounds in combating pathogenic stressors through oxidative mechanisms [20], a principle applicable to dysmenorrhea management through antioxidant plant extracts.

This study aimed to: (1) conduct an ethnobotanical survey to document plant species traditionally used for menstrual pain treatment in Dekina LGA; (2) perform comprehensive phytochemical profiling of the most frequently cited species; and (3) evaluate their antioxidant potentials through validated in vitro assays. These objectives align with the growing need to bridge traditional knowledge with contemporary scientific validation and support the development of evidence-based phytotherapeutics for dysmenorrhea management.

MATERIALS AND METHODS

Study Area: This research was conducted in Dekina LGA (9.3°N, 7.5°E), Kogi State, Nigeria. The region has a tropical savanna climate with a mean annual rainfall of 1,000–1,500 mm and a predominantly agrarian population. The study was approved by the Research Ethics Committee of the Department of Plant Science and Biotechnology, Prince Abubakar Audu University (PAAU), Anyigba, Nigeria (approval reference: PAAU/REC/2023/08).

Ethnobotanical Survey: Ethnobotanical surveys were conducted between March and August 2023 using semi-structured questionnaires and focus group discussions with 45 traditional healers, herbalists, and community health workers. Informed written consent was obtained from all participants prior to interviews. Data collected included plant species, local names, plant parts used, preparation methods, dosage, duration of use, and indications for menstrual pain treatment. Plant voucher specimens were collected, botanically identified using standard taxonomic keys, authenticated by the Department herbarium at PAAU, and deposited for reference with accession numbers assigned.

Plant Material Collection and Preparation: Five plant species most frequently cited for menstrual pain treatment (≥60% respondent consensus) were selected for laboratory analysis. These included *Hibiscus sabdariffa* L. (Malvaceae, local name: zobo), *Ocimum basilicum* L. (Lamiaceae, local name: scent leaf), *Zingiber officinale* Roscoe (Zingiberaceae, local name: ginger), *Mentha spicata* L. (Lamiaceae, local name: mint), and *Allium sativum* L. (Amaryllidaceae, local name: garlic). Plant materials (fresh leaves for H. sabdariffa, O. basilicum, and M. spicata; fresh rhizomes for Z. officinale; fresh bulbs for A. sativum) were collected during peak growing season, washed with distilled water, shade-dried at 25°C for 14 days, and ground into fine powder (1 mm mesh) using an electric mill.

Extract Preparation: Aqueous extracts were prepared by decoction, replicating the traditional preparation method used by respondents. Fifty grams of each plant powder was boiled in 500 mL of distilled water for 30 minutes, cooled to room temperature, filtered through Whatman filter paper (No. 1), and the filtrate was lyophilized (Labconco FreeZone 12, Kansas City, MO, USA) to obtain dried extract powder. Lyophilized extracts were stored at 4°C in opaque containers prior to analysis.

Phytochemical Analysis: Preliminary phytochemical screening was performed using standard qualitative methods to detect alkaloids (Mayer's test), flavonoids (aluminum chloride test), tannins (ferric chloride test), saponins (foam test), and glycosides (Keller-Killiani test)[8]. Tests were performed in triplicate and results recorded as present (+) or absent (−).

Determination of Total Phenolic Content: Total phenolic content was determined using the Folin-Ciocalteu colorimetric method. Extract solutions (50 µL, 1 mg/mL in distilled water) were mixed with Folin-Ciocalteu reagent (250 µL, 1:10 dilution) and distilled water (700 µL). After 5 minutes, sodium carbonate solution (1 mL, 7.5% w/v) was added, and the mixture was incubated in the dark at room temperature for 30 minutes. Absorbance was measured at 765 nm using a spectrophotometer (UV-VIS Spectrophotometer, Jenway 6300, Essex, UK). Gallic acid (0–100 µg/mL) was used as standard. Results were expressed as milligrams of gallic acid equivalents per 100 g dry weight (mg GAE/100g DW). All measurements were performed in triplicate.

DPPH Radical Scavenging Activity: 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity was assessed according to established protocols [9]. Extract solutions (0–200 µg/mL in methanol) were mixed with DPPH solution (100 µM in methanol) in equal volumes and incubated in the dark at room temperature for 30 minutes. Absorbance was measured at 517 nm. Ascorbic acid (0–200 µg/mL) was used as a positive control. The percentage of radical scavenging activity was calculated as: $[(A_o - A_1)/A_o] \times 100$, where A_o is absorbance of DPPH control and A_1 is absorbance of sample. IC_{50} values (concentration inhibiting 50% of DPPH) were determined from dose-response curves. All analyses were performed in triplicate.

Ferric Reducing Antioxidant Power (FRAP) Assay: Ferric Reducing Antioxidant Power (FRAP) assay was conducted

as previously described [10]. FRAP working reagent was prepared fresh by mixing acetate buffer (300 mM, pH 3.6), 2,4,6-tripyridyl-s-triazine (10 mM in HCl 40 mM), and ferric chloride (20 mM) in a ratio of 10:1:1. Extract solutions (10 µL) were added to FRAP reagent (190 µL) and incubated at 37°C for 30 minutes. Absorbance was measured at 593 nm against a blank. Ferrous sulfate (0–1000 µM) was used as standard. Results were expressed as millimoles of ferrous sulfate equivalents per 100 g dry weight (mmol ferrous sulfate equivalents [FSE]/100g DW). All measurements were performed in triplicate.

Statistical Analysis: Data were presented as mean ± standard deviation (SD) of three replicate measurements. Differences between samples were compared using one-way ANOVA followed by Duncan's multiple-range test. Correlation analysis was performed between total phenolic content and antioxidant activities. Statistical significance was set at $p < 0.05$. Data analysis was performed using IBM SPSS Statistics version 26 (Armonk, NY, USA).

RESULTS

Ethnobotanical Survey Findings: The ethnobotanical survey documented 23 plant species from 19 botanical families traditionally used for menstrual pain treatment in Dekina LGA. Five species showed the highest respondent consensus (≥60%) and were selected for further analysis. Plant parts commonly used were leaves (65%), rhizomes/bulbs (20%), and whole plant (15%). Primary preparation methods reported were decoction (75%), maceration (15%), and fresh juice extraction (10%). Dosage frequency ranged from once daily to three times daily, with treatment duration typically extending throughout or a few days before menstruation.

Phytochemical Composition: Qualitative phytochemical screening results for the five plant species are presented in Table 1. All five species contained significant levels of alkaloids, flavonoids, tannins, and saponins. Alkaloids were present in all species, being most abundant in *A. sativum* and *Z. officinale*. Flavonoids were detected in all species, with particularly strong reactions in *H. sabdariffa* and *O. basilicum*. Tannins were present in all species, with the highest concentrations observed in *M. spicata*. Glycosides were detected in *H. sabdariffa*, *Z. officinale*, and *M. spicata*.

Table 1. Phytochemical Composition of Selected Medicinal Plants Used for Menstrual Pain Treatment in Dekina LGA

Plant Species	Alkaloids	Flavonoids	Tannins	Saponins	Glycosides
Hibiscus sabdariffa	+	++	++	+	+
Ocimum basilicum	+	++	+	+	−
Zingiber officinale	++	+	++	+	+
Mentha spicata	+	+	++	+	+
Allium sativum	++	+	+	+	−

++ = strong presence; + = moderate presence; − = absent

Total Phenolic Content: Results for total phenolic content determination are presented in Table 2. *Hibiscus sabdariffa* demonstrated the highest total phenolic content at 245.32±8.21 mg gallic acid equivalents (GAE)/100g DW, followed by *Mentha spicata* (198.45±6.87 mg GAE/100g dry weight (DW)) and *Ocimum basilicum* (167.89±5.42 mg GAE/100g DW). *Zingiber officinale* and *Allium sativum* showed lower but still significant phenolic contents at 142.56±4.95 and 128.73±4.18 mg GAE/100g DW, respectively.

Table 2. Total Phenolic Content of Plant Extracts (mg GAE/100g Dry Weight)

Plant Species	Total Phenolic Content (mg GAE/100g DW)
Hibiscus sabdariffa	245.32 ± 8.21[a]
Mentha spicata	198.45 ± 6.87[b]
Ocimum basilicum	167.89 ± 5.42[c]
Zingiber officinale	142.56 ± 4.95[d]
Allium sativum	128.73 ± 4.18[d]

Values are expressed as mean ± SD (n = 3). Different superscript letters indicate statistically significant differences ($p < 0.05$) among samples, as determined by Duncan's multiple-range test.

Antioxidant Activity: Results for DPPH radical scavenging and FRAP assay are presented in Table 3. *Zingiber officinale* exhibited the strongest DPPH radical scavenging activity with IC_{50} value of 38.45±2.15 µg/mL, followed by *Mentha spicata* (IC_{50} = 52.34±2.89 µg/mL) and *Hibiscus sabdariffa* (IC_{50} = 63.21±3.12 µg/mL). *Ocimum basilicum* and *Allium sativum* showed moderate DPPH scavenging activity with IC_{50} values of 81.67±3.95 and 95.43±4.62 µg/mL, respectively. Ascorbic acid (positive control) showed IC_{50} value of 18.34±1.05 µg/mL.

In the FRAP assay, *Hibiscus sabdariffa* demonstrated the highest reducing power at 156.78±7.23 mmol FSE/100g DW, while *Mentha spicata* (134.56±6.12 mmol FSE/100g DW), *Zingiber officinale* (118.43±5.67 mmol FSE/100g DW), *Ocimum basilicum* (96.34±4.89 mmol FSE/100g DW), and *Allium sativum* (82.15±3.98 mmol FSE/100g DW) showed progressively lower values. Ferrous sulfate standard showed FRAP value of 989.23±45.12 mmol FSE/100g.

Pearson correlation analysis demonstrated significant positive correlation between total phenolic content and both DPPH scavenging activity ($r = 0.89$, $p < 0.01$) and FRAP values ($r = 0.92$, $p < 0.01$), indicating that phenolic compounds are primary contributors to the antioxidant potential of these extracts.

Table 3. Antioxidant Activity of Plant Extracts: DPPH Scavenging and FRAP Assay Results

Plant Species	DPPH IC_{50} (µg/mL)	FRAP (mmol FSE/100g DW)
Zingiber officinale	38.45 ± 2.15[a]	118.43 ± 5.67[b]
Mentha spicata	52.34 ± 2.89[b]	134.56 ± 6.12[a]
Hibiscus sabdariffa	63.21 ± 3.12[c]	156.78 ± 7.23[c]
Ocimum basilicum	81.67 ± 3.95[d]	96.34 ± 4.89[d]
Allium sativum	95.43 ± 4.62[e]	82.15 ± 3.98[e]
Ascorbic acid (positive control)	18.34 ± 1.05	–
Ferrous sulfate (positive control)	–	989.23 ± 45.12

Values are expressed as mean ± SD (n = 3). Different superscript letters indicate statistically significant differences ($p < 0.05$) among samples, as determined by Duncan's multiple-range test.

DISSCUSSION

This study represents one of the first comprehensive integrations of ethnobotanical survey data with phytochemical and antioxidant analyses for medicinal plants used in menstrual pain management in Dekina LGA, Kogi State, Nigeria. The ethnobotanical survey documented 23 plant species, highlighting the region's

rich traditional knowledge of plant-based therapeutics for dysmenorrhea management.

The phytochemical analysis revealed that all five selected species contain multiple bioactive compound classes, particularly alkaloids, flavonoids, tannins, and saponins. These compounds have well-established roles in anti-inflammatory, antispasmodic, and analgesic activities relevant to dysmenorrhea pathophysiology. Alkaloids possess analgesic properties through multiple mechanisms, including inhibition of prostaglandin synthesis and opioid receptor interactions [11]. Flavonoids exhibit potent anti-inflammatory effects through NF-κB pathway inhibition and cytokine suppression [12]. Tannins exhibit astringent properties and may reduce uterine bleeding intensity through vascular contraction mechanisms [13].

At the compound level, the observed bioactivities can be attributed to specific phytochemicals characteristic of each species. In *H. sabdariffa*, hibiscus acid, protocatechuic acid, and anthocyanins (delphinidin-3-sambubioside, cyanidin-3-sambubioside) are the principal polyphenolics responsible for the high total phenolic content and strong ferric reducing capacity [21]. These compounds exert anti-inflammatory effects by inhibiting cyclooxygenase (COX-1 and COX-2), directly suppressing prostaglandin E2 synthesis and thereby attenuating uterine contractility in dysmenorrhea. In *Z. officinale*, the superior DPPH radical scavenging activity (concentration required to scavenge 50% of DPPH radicals [IC50] = 38.45 +/- 2.15 ug/mL) is attributable primarily to [6]-gingerol, [6]-shogaol, and zingerone, phenolic ketones known to inhibit both COX and lipoxygenase (LOX) pathways, reduce prostaglandin and leukotriene biosynthesis, and scavenge reactive oxygen species through hydrogen atom transfer mechanisms [22]. In *M. spicata*, rosmarinic acid and luteolin are the principal anti-inflammatory constituents, with rosmarinic acid demonstrated to inhibit complement activation and arachidonic acid metabolite formation, contributing to both the high phenolic content and potent FRAP value observed. In *O. basilicum*, eugenol and linalool confer spasmolytic and analgesic activity by relaxing smooth muscle through calcium channel antagonism, which may directly reduce the uterine hypercontractility characteristic of primary dysmenorrhea. Collectively, these compound-level insights reveal that the observed antioxidant and anti-inflammatory activities are mechanistically relevant to dysmenorrhea pathophysiology and are not merely incidental to phytochemical richness [23].

The significantly elevated total phenolic content in *H. sabdariffa* (245.32±8.21 mg GAE/100g DW) aligns with previous reports documenting the high polyphenolic profile of hibiscus species [14]. The strong DPPH radical scavenging activity of *Z. officinale* (IC_{50} = 38.45±2.15 µg/mL) supports extensive literature demonstrating the potent antioxidant properties of gingerol and shogaol compounds found in ginger rhizomes [15]. These antioxidant activities are mechanistically relevant to dysmenorrhea management, as oxidative stress suppression reduces activation of the prostaglandin-mediated inflammatory cascade and alleviates uterine pain perception. Biologically, the IC50 values obtained for *Z. officinale* and *M. spicata* in this study are comparable to or lower than those reported for aqueous plant extracts used in menstrual pain management in other African settings, where IC50 values typically range from 40-120 ug/mL for DPPH scavenging, thereby confirming the competitive antioxidant potency of these Dekina species [22]. The FRAP values of *H. sabdariffa* (156.78 +/- 7.23 mmol FSE/100g DW) substantially exceed those reported for several commercially valorized antioxidant plants, underscoring its potential as a functional food ingredient for managing oxidative stress-associated conditions, including dysmenorrhea. Within a functional food science framework, these findings are consistent with the concept that bioactive compounds in whole food matrices can modulate biological processes through

complementary antioxidant and anti-inflammatory mechanisms, as elaborated in recent translational frameworks for functional food science [24].

The positive correlations between total phenolic content and antioxidant activities ($r = 0.89–0.92$, $p < 0.01$) provide evidence that phenolic compounds serve as the primary antioxidant contributors in these plant extracts. This finding supports the use of total phenolic content as a reliable proxy for estimating antioxidant potential in traditional medicinal plants, consistent with observations in other functional food systems from the region [16,19]. The integration of bioactive compound assessment with traditional knowledge documentation represents an important approach to validating indigenous food and medicinal practices [17], particularly in resource-limited settings where functional foods offer accessible opportunities for health promotion.

Scientific Innovations: This research provides novel insights into the phytochemical basis for the traditional use of five medicinal plants in dysmenorrhea management within the Dekina region. By establishing quantitative relationships between bioactive compounds and antioxidant potentials, the study advances understanding of mechanisms underlying traditional therapeutic practices. The identification of specific polyphenolic profiles in each species enables targeted development of functional food ingredients and phytopharmaceutical formulations optimized for dysmenorrhea management.

Practical Implications: These findings support the development of evidence-based herbal formulations combining species with complementary bioactive profiles—for example, integrating *Z. officinale* (strong antioxidant and analgesic properties) with *H. sabdariffa* (high phenolic content and reducing power) to create synergistic therapeutic formulations. Such approaches could provide accessible, affordable alternatives to conventional NSAIDs, particularly for populations in resource-limited settings. Furthermore, the documented phytochemical profiles provide baselines for quality control standardization in traditional medicine production and enable regulatory pathways for phytotherapeutic product development [18].

Importantly, the translation of these plant-based candidates into standardized functional food or nutraceutical products should be guided by the Functional Food Center (FFC)'s 17-step process for achieving functional food status [24]. This structured framework — encompassing steps from initial identification of bioactive compounds and safety profiling, through preclinical and clinical validation, to labeling, regulatory approval, and post-market surveillance — provides a scientifically rigorous and regulatory-compliant pathway for developing the herbal combinations identified in this study into consumer-ready functional food products. Applying this framework to *Z. officinale*, *H. sabdariffa*, and the other documented species would systematically address the evidence gaps currently limiting their clinical translation, including bioavailability assessment, effective dosage determination, and population-specific efficacy validation. Recent advances in characterizing bioactive compounds from traditionally fermented foods and medicinal plants in Nigeria have established frameworks for linking chemical composition with functional properties, further supporting the development of region-specific functional food applications aligned with the FFC model [19–20].

The antioxidant activities measured in this study suggest potential benefits for managing oxidative stress-associated dysmenorrhea symptoms; however, clinical efficacy must be demonstrated through randomized controlled trials. Future research should investigate the mechanisms of pain alleviation in cell culture and animal models, establish optimal dosage formulations consistent with the FFC's dosage characterization steps, and conduct clinical trials to confirm therapeutic efficacy

and safety profiles in dysmenorrheic populations — all integral components of the FFC's 17-step validation continuum.

CONCLUSION

This study successfully documented, chemically profiled, and evaluated the antioxidant potential of five medicinal plants traditionally used for menstrual pain treatment in Dekina LGA, Kogi State, Nigeria. All analyzed species demonstrated significant levels of bioactive compounds and antioxidant activities, with *H. sabdariffa* showing the highest phenolic content and reducing power, while *Z. officinale* exhibited the strongest DPPH radical scavenging activity. Strong correlations between total phenolic content and antioxidant activities indicate that phenolic compounds are key mediators of the observed bioactivities. These findings provide scientific validation for the traditional use of these plants and establish a foundation for the development of evidence-based phytotherapeutics for dysmenorrhea management. Further pharmacological studies and clinical trials are recommended to establish efficacy, safety profiles, and optimal therapeutic formulations.

List of Abbreviations: DPPH, 1,1-diphenyl-2-picrylhydrazyl; FRAP, Ferric reducing antioxidant power; TPC, Total phenolic content; NSAIDs, Nonsteroidal anti-inflammatory drugs; ROS: Reactive oxygen species; LGA: Local government area; IC_{50}: Inhibitory concentration 50; mg GAE/100g DW, Milligrams of gallic acid equivalents per 100 g dry weight; mmol FSE/100g DW, Millimoles of ferrous sulfate equivalents per 100 g dry weight

Authors' Contributions: H.A.O. (Department of Engineering and Space Systems, NASRDA) conceptualized and designed the study, oversaw systems reliability aspects of the research methodology, performed statistical data analysis, and drafted the manuscript. A.D.O. (Department of Plant Science and Biotechnology, PAAU) conducted the ethnobotanical surveys, coordinated plant material collection and botanical identification, authenticated voucher specimens at the PAAU herbarium, and contributed to manuscript revision. M.T.B. (Department of Plant Science and Biotechnology, PAAU) performed all phytochemical screening and antioxidant assays (DPPH, FRAP, and total phenolic content determination), compiled and interpreted laboratory data, and supervised the experimental work as corresponding author. Z.A.D. (Department of Microbiology, PAAU) contributed to microbiological and biochemical data interpretation, assisted in the contextualization of findings within functional food and bioactive compound frameworks, and reviewed and revised the manuscript for intellectual content. All authors read and approved of the final manuscript.

Competing Interests: The authors declare no competing interests.

Acknowledgment and Funding: The authors are grateful to all traditional healers and community members in Dekina LGA who shared their ethnobotanical knowledge. We acknowledge the Department of Plant Science and Biotechnology, Prince Abubakar Audu University, Anyigba, Nigeria, for providing laboratory facilities. This research received no specific grant from any funding agency.

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