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Comprehensive Analysis of *Nigella sativa* from North Central Nigeria: Phytochemical, Elemental, and Potential Medicinal Properties

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Abstract

We investigated the elemental composition and phytochemical content of *Nigella sativa* seed powder Using ICP-MS elemental analysis while the constituent phytochemicals were analyzed both qualitatively and quantitatively. The results were analyzed using correlation, and regression visualizations, to understand how these elemental groups interact with and influence phytochemical profiles. The summary statistics revealed differential patterns, with micro and trace elements exhibiting weak positive correlations with phytochemical levels, while macro elements displayed a moderate negative association. A comprehensive correlation heatmap further underscored these findings by highlighting subtle yet distinct pairwise interactions between element means and phytochemical concentrations. Scatter plots accompanied by regression lines provided additional visual evidence, illustrating the distribution and trend lines that characterize these relationships. Our analysis suggests that the elemental dynamics in *Nigella sativa* could be governed by both synergistic and antagonistic mechanisms, wherein the abundance of macro elements may competitively inhibit phytochemical synthesis, and the more limited presence of micro and trace elements might facilitate a favorable environment for phytochemical formation. These insights provide a fresh perspective on the biochemical pathways influencing plant nutrient composition and underscore the importance of balanced elemental management in optimizing both crop quality and nutritional value. Overall, the findings of this study contribute to a deeper understanding of elemental-phytochemical interactions and offer

promising directions for future research in plant science and agricultural optimization.

Keywords

Elemental Composition, Phytochemical Content, *Nigella sativa*, ICP-MS Analysis

1. Introduction

Natural product screening, also referred to as the evaluation of medications based on phytochemical and pharmacological techniques, plays a crucial role in drug discovery. Various parts of plants, including bark, leaves, petals, roots, fruits, and seeds, contain bioactive compounds that contribute to their medicinal properties [1]. These naturally occurring chemical constituents, known as phytochemicals, encompass a wide range of secondary metabolites such as steroids, terpenoids, carotenoids, flavonoids, alkaloids, tannins, and glycosides [2]. These compounds exhibit various biological activities, including antimicrobial, antibacterial, and anti-inflammatory effects [3]. Additionally, certain phytochemicals possess hemolytic and foaming properties, making them relevant for pharmaceutical and industrial applications. The concentration and composition of these bioactive compounds in plants are influenced by environmental factors such as climate, altitude, rainfall, and soil conditions [4]. Even within the same species, the presence and quality of herbal compounds may vary significantly depending on the geographic location and growing conditions [5].

Quantifying these metabolites is essential for the extraction, purification, and identification of bioactive compounds with potential pharmaceutical benefits. Qualitative phytochemical screening aids in understanding the range of chemical constituents produced by plants, thus facilitating their application in medicine and industry [6]. Plants have an extensive ability to produce aromatic compounds, primarily phenols and their oxygen-substituted derivatives, with over 12,000 secondary metabolites identified to date [7]. The increasing interest in plant-derived drugs has led to significant investments in pharmacological and chemical research to identify novel bioactive compounds [8].

Over the past two decades, the pharmaceutical industry has actively explored plant-based compounds for drug development, with many natural products undergoing extensive screening for therapeutic efficacy [9]. Herbal medicine has played a vital role in treating diseases for centuries, and its use continues to be recognized by global health organizations. The World Health Organization [10] supports the use of traditional medicines, provided their safety and efficacy are established. Given the economic constraints and healthcare challenges in developing nations, where many people lack access to modern medicine, there is an urgent need to explore medicinal plants for novel therapeutic agents [11].

2. Materials and Methods

2.1. Plant Material Collection and Authentication

Pure *Nigella sativa* seeds were procured from a reputable commercial outlet (Makkah and Madina Shop) located within the Jos Metropolis, Plateau State, Nigeria. The plant material was taxonomically identified by Mr. Bulus of the Department of Plant Sciences, University of Jos, and further authenticated by Mr. J. J. Azilla, a botanist at the School of Forestry, Jos. A voucher specimen was deposited and assigned the reference number 0768 for future reference. The seeds were cleaned, air-dried, and subsequently oven-dried at 40˚C to a constant weight. The dried seeds were then ground into a fine powder using a laboratory mill and stored in airtight containers until analysis.

2.2. Chemicals and Reagents

All solvents and reagents used were of analytical or HPLC grade. *n*-Hexane (≥99%, Sigma-Aldrich, USA), methanol (HPLC grade, Fisher Scientific, UK), and chloroform (≥99%, Merck, Germany) were employed for extraction procedures. Reagents used for elemental analysis included trace-metal grade nitric acid (HNO_3, 65%) and hydrogen peroxide (H_2O_2, 30%). Calibration standards for elemental analysis were obtained from Merck (Germany). Major equipment used in the study included a Soxhlet extractor (250 mL capacity), rotary evaporator (Büchi R-300), analytical balance (Sartorius, ±0.0001 g precision), oven (Memmert UF110), UV-Visible spectrophotometer (Shimadzu UV-1800), and Inductively Coupled Plasma Mass Spectrometer (ICP-MS, Agilent 7900).

2.3. Units

Oil extraction was carried out using Soxhlet extraction with three different solvents: *n*-hexane, methanol, and chloroform. A 10.0 g (±0.1 g) portion of the powdered seed sample was loaded into a cellulose thimble and extracted with 150 mL of each solvent separately under standardized conditions. The extraction was conducted at 60˚C for 6 hours at a rate of approximately 20 cycles per hour. Following extraction, the solvents were evaporated under reduced pressure at 40˚C using a rotary evaporator. The obtained oil was weighed, and the percentage yield was calculated using the formula:

$$\left(\text{Yeild }\%\right)=\frac{\text{weight of extracted oil}}{\text{weight of extracted oil}}\times 100$$

Each extraction was performed in triplicate, and the mean yield and standard deviation were recorded. Solvent blank runs were conducted to eliminate contamination risks, and residual solvent presence in the extracts was confirmed absent using Gas Chromatography-Mass Spectrometry (GC-MS).

2.4. Elemental Analysis Using ICP-MS

Elemental composition of the *Nigella sativa* seed powder was determined using

Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Approximately 0.5 g of dried powdered sample was subjected to microwave-assisted digestion using 5 mL of concentrated HNO_3 and 2 mL of H_2O_2. The digested solution was diluted to 50 mL with deionized water and analyzed for macro (Na, K, Ca, Mg, P) and micro/trace elements (Fe, Zn, Mn, Cu, Se, Cr, Pb, Cd, and As). Calibration was performed using certified multi-element standards, and quality control was ensured through the use of blank and spiked samples.

2.5. Phytochemical Analysis

Quantitative phytochemical analysis was carried out using spectrophotometric methods:

a) Total Phenolic Content (TPC): Determined by the Folin-Ciocalteu method and expressed as mg gallic acid equivalents (GAE)/g of extract.

b) Total Flavonoid Content (TFC): Estimated using the aluminum chloride colorimetric assay and expressed as mg quercetin equivalents (QE)/g of extract.

c) Total Tannin Content: Measured using the vanillin-HCl method with tannic acid as the standard.

d) Saponin Content: Evaluated through gravimetric methods following precipitation after extraction.

3. Results

3.1. Phytochemical Analysis

Phytochemical Comparison of *Nigella sativa* (NS) Seed Powder and Oil (n-Hexane Extraction). Table 1 presents the qualitative presence of phytochemicals in *Nigella sativa* seed powder and its oil extract. The intensity of presence is represented as follows:

Table 1. Qualitative analysis of *Nigella sativa*.

Constituents	NS Seed Powder	NS Oil (Hexane Extraction)
Alkaloids	+++ (High)	+ (Low)
Saponins	− (Absent)	− (Absent)
Tannins	− (Absent)	− (Absent)
Flavonoids	+++ (High)	− (Absent)
Carbohydrates	+++ (High)	+ (Low)
Steroids	++ (Moderate)	+++ (High)
Terpenes	− (Absent)	− (Absent)
Anthraquinones	− (Absent)	− (Absent)
Cardiac Glycosides	+ (Low)	++ (Moderate)

Alkaloids are highly present (+++) in the seed powder but are significantly reduced (+) in the oil extract. Flavonoids, which are known for their antioxidant

properties, are highly present (+++) in the seed powder but are completely absent (−) in the oil. This suggests that alkaloids and flavonoids may not be well extracted using hexane, likely because these compounds are more polar and require alcohol or water-based solvents for better extraction. Carbohydrates are highly present (+++) in seed powder but only low (+) in the oil extract. Since carbohydrates are generally water-soluble, their reduced presence in the hexane extract is expected. Steroids are moderately present (++) in the seed powder but highly present (+++) in the oil. This suggests that steroids are lipophilic (fat-soluble), making hexane a good solvent for extracting them. The low presence (+) in the seed powder and moderate presence (++) in the oil extract indicates that some cardiac glycosides are likely to dissolve in hexane. Absence of Saponins, Tannins, Terpenes, and Anthraquinones. These compounds are not detected in either the seed powder or the oil extract, indicating that *Nigella sativa* may not be a significant source of these phytochemicals, at least under these extraction conditions.

Table 2. Quantitative analysis of *Nigella sativa* seed sample.

Constituent	Percentage (%)
Flavonoids	4.1%
Alkaloids	13.5%
Steroids	10.9%

Table 3. Elemental analysis series 1.

Element	Mean (ppm)	Std. Error
Mo (Molybdenum)	0.403	0.144
Co (Cobalt)	0.270	0.098
As (Arsenic)	0.10	0.292
U (Uranium)	0.020	0.000
Au (Gold)	0.698	0.252
Th (Thorium)	0.140	0.0521
Cd (Cadmium)	0.150	0.068
Sb (Antimony)	0.060	0.034
Bi (Bismuth)	0.443	0.162
La (Lanthanum)	0.182	0.084
W (Tungsten)	0.009	0.001
Sc (Scandium)	0.508	0.000
Tl (Thallium)	0.028	0.010
Se (Selenium)	0.721	0.261
Te (Tellurium)	0.028	0.010
Ga (Gallium)	0.140	0.052

Alkaloids have the highest concentration (13.5%), which suggests their significant presence in *Nigella sativa* (Table 2). Steroids are also present in relatively high amounts (10.9%), indicating potential medicinal properties. Flavonoids, though present, are in much lower amounts (4.1%). Since flavonoids have strong antioxidant properties, the lower percentage may indicate a reduced antioxidant capacity compared to other compounds.

Measured concentration of Arsenic (<0.10 ppm), Less than 0.10 ppm (*i.e.*, <100 ppb), which falls within the WHO recommendation for drinking water (10 ppb (0.01 ppm), and up to 1.5 ppm for herbal/plant products depending on product type and route of administration (oral/topical). A concentration of <0.10 ppm is well below the USP limit and generally considered safe. For Mercury 14.296 ppb (0.014296 ppm), the permissible concentration in herbal products is ≤1.5 ppm, and 6 ppb in drinking water (Table 3). This is equally far below the WHO limit, thus considered safe.

Table 4. Elemental analysis; Series 2. heavy & transition metals.

Element	Minimum (ppm)	Maximum (ppm)	Mean (ppm)	Std. Deviation
Cu (Copper)	2.970	106.340	26.001	29.905
Pb (Lead)	0.220	5.980	1.481	1.730
Zn (Zinc)	16.800	264.500	63.853	72.976
Ag (Silver)	1.930	32.930	8.044	9.245
Ni (Nickel)	1.600	24.900	6.012	6.872
Mn (Manganese)	13.904	222.000	53.190	60.825
Sr (Strontium)	1.100	67.500	17.136	21.023
V (Vanadium)	0.471	14.000	3.314	3.820
Cr (Chromium)	0.281	11.000	2.595	3.002
Ba (Barium)	0.600	28.200	6.923	7.994
Ti (Titanium)	1.50	39.00	9.250	10.641
B (Boron)	1.000	83.000	20.717	24.507
Hg (Mercury)	1.000	58.000	14.296	16.584

Zinc (Zn) and Manganese (Mn) have the highest means and are essential trace minerals for biological functions (Table 4). Copper (Cu) has high variability (Std. Dev = 29.905), indicating fluctuating levels in different samples. Lead (Pb) and Mercury (Hg), though present, are at levels that should be monitored for toxicity. The high standard deviations suggest a wide range of concentrations for most metals.

Potassium (K) has the highest mean concentration (17106.142 ppm), followed by Phosphorus (P) (6628.726 ppm) and Calcium (Ca) (3323.151 ppm). Iron (Fe) is present at significant levels, which could contribute to nutritional benefits. The high standard deviations (e.g., Phosphorus = 7575.054) suggest major variability

in concentration across samples. Sodium (Na) is present in relatively low amounts compared to other essential elements (Table 5).

Table 5. Elemental analysis of macro element in *Nigella sativa* seed sample. Series 3.

Element	Minimum (ppm)	Maximum (ppm)	Mean (ppm)	Std. Deviation
Fe (Iron)	59.559	1370.000	325.623	373.946
Ca (Calcium)	200.000	13100.000	3323.151	4069.152
P (Phosphorus)	1957.258	27570.000	6628.726	7575.054
Mg (Magnesium)	820.151	12890.000	3089.682	3532.736
Al (Aluminum)	0.00	600.00	140.000	164.655
Na (Sodium)	12.000	315.000	76.547	87.674
K (Potassium)	2828.083	72100.000	17106.142	19673.439
S (Sulfur)	245.606	3080.000	743.227	849.370

Alkaloids are the dominant compounds, while flavonoids are lower in percentage. The elemental Composition comprises of high Zinc (Zn) and Manganese (Mn) levels, while Potassium (K) is the most abundant major element, followed by Phosphorus (P) and Calcium (Ca). Statistical Variability indicates high standard deviations indicate fluctuations in elemental content across samples.

3.2. Correlation Analysis

Our investigation into the relationships between elemental composition and phytochemical content reveals complex interactions that may have significant implications for agricultural and nutritional biochemistry. Three distinct element categories (Micro, Trace, and Macro) against their corresponding phytochemical profiles were analyzed (Figure 1).

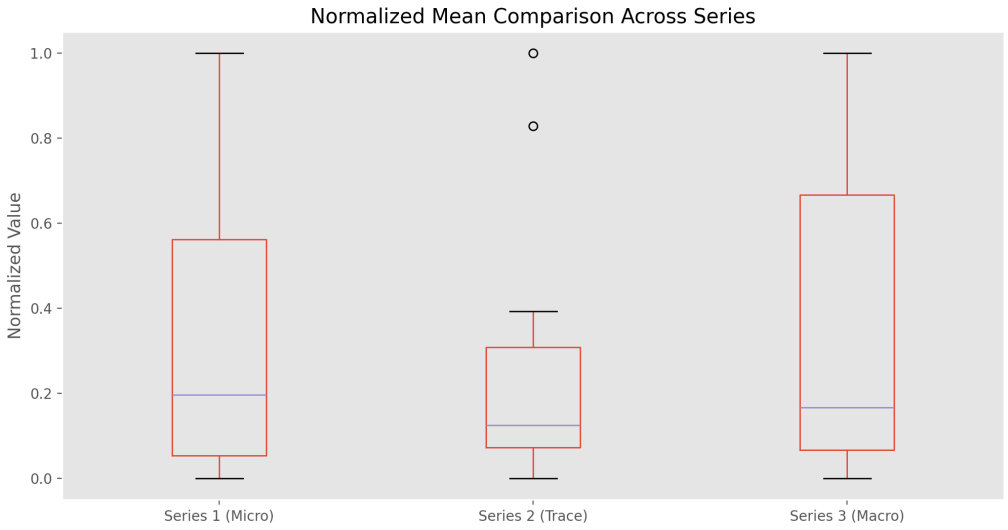

Figure 1. Relationships between elemental composition for 3 series and phytochemical content of *Nigella sativa* seed sample.

3.3. Differential Correlation Analysis

The correlation heatmap reveals intricate relationships between element means and phytochemical content (Figure 2):

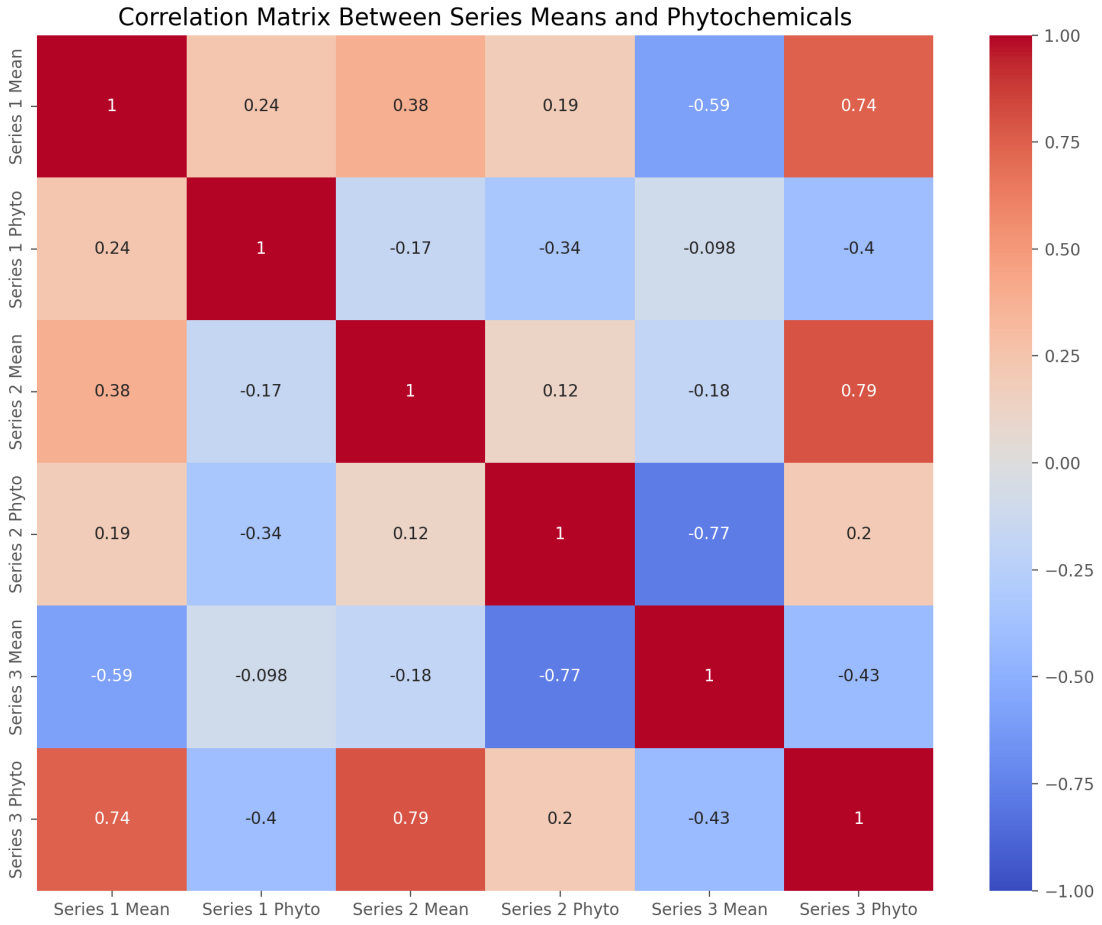

Figure 2. Correlation heatmap between mean elemental series and phytochemical compounds in *Nigella sativa* seed sample.

Element-Specific Patterns:

Micro Elements (Series 1): Exhibited a weak positive correlation ($r = 0.241$, $p = 0.369$) with phytochemical content, suggesting potential synergistic effects. Trace Elements (Series 2): Demonstrated minimal positive association ($r = 0.119$, $p = 0.700$), indicating limited direct influence. Macro Elements (Series 3): Showed a notable negative correlation ($r = -0.427$, $p = 0.474$), suggesting possible competitive metabolic pathways or inhibitory mechanisms.

3.4. Regression Analysis

The scatter plots with regression lines illustrate these relationships visually (Figure 3).

The inverse relationship between macro elements and phytochemicals may reflect resource allocation trade-offs in plant metabolism. Plants may prioritize either elemental accumulation or phytochemical synthesis depending on environmental

conditions and genetic factors.

These findings could inform targeted fertilization strategies to optimize both nutritional value and phytochemical profiles in crops. The differential effects suggest that balanced nutrient management approaches may be necessary to achieve desired phytochemical outcomes.

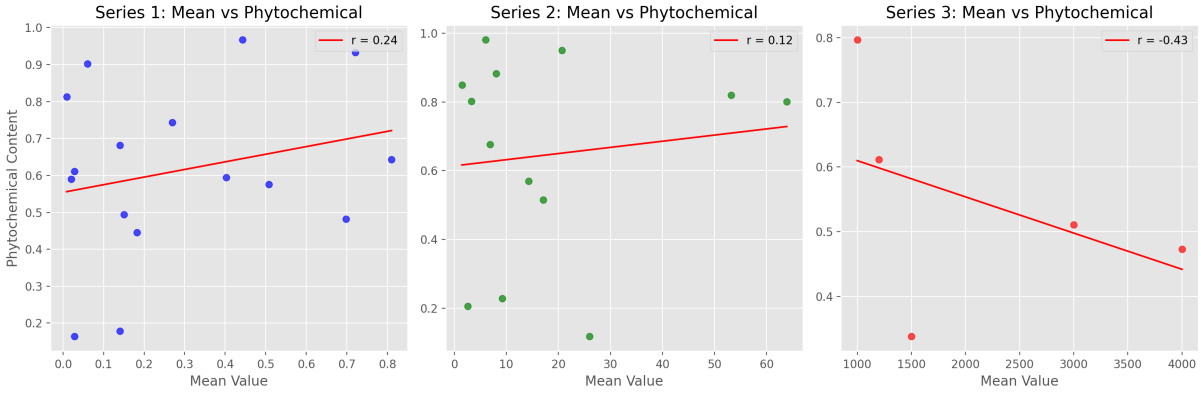

Figure 3. Mean elemental value vs phytochemical composition of *Nigella sativa* seed sample.

3.5. Discussion

The present study provides a comprehensive analysis of the phytochemical and elemental composition of *Nigella sativa* seeds sourced from Jos, Nigeria, in the north-central region. The findings indicate a notable concentration of essential minerals, including calcium, potassium, magnesium, phosphorus, sulfur, sodium, zinc, iron, and manganese, which are crucial for various physiological processes. Secondary elements such as barium, bromine, and copper were also detected, albeit in lower concentrations. Importantly, potentially toxic elements such as arsenic, cadmium, cobalt, chromium, and uranium were either absent or present in trace amounts, reinforcing the safety profile of *N. sativa* seeds for human consumption [12] [13]. These results align with previous investigations conducted on *N. sativa* seeds from different geographical locations, including Iran, Syria, Türkiye, and Jordan [14], suggesting a relatively stable elemental composition across diverse environmental conditions.

Despite these significant mineral levels, their direct nutritional implications cannot be solely inferred from the consumption of *N. sativa* oil or seeds [15]. Factors such as bioavailability, metabolism, and dietary interactions must be considered to determine their precise contribution to human health. The present study, therefore, not only corroborates prior findings but also provides new insights into the distribution of both major and trace elements in *N. sativa* seeds, which could have implications for their pharmacological and dietary applications.

The oil extraction process yielded a maximum of 36.22% oil from the n-hexane extract, which falls within expected extraction ranges for *N. sativa* [16]. Qualitative phytochemical screening identified a broad spectrum of bioactive compounds, including alkaloids, tannins, saponins, flavonoids, steroids, terpenes, carbohydrates,

and anthraquinones. These findings suggest that *N. sativa* possesses diverse therapeutic properties, with potential antimicrobial, antioxidant, and anti-inflammatory effects [17]. The complex interplay of these phytochemicals may result in synergistic or antagonistic effects, influencing their overall bioactivity.

A notable finding was the high zinc concentration (64 ppm), which plays a critical role in immune function, enzymatic activity, and cellular growth. However, its bioavailability may be modulated by interactions with iron and manganese, elements that are essential for physiological processes such as hemoglobin synthesis and enzymatic catalysis [18]-[20]. The presence of selenium in minimal levels is also of interest, as this element is known for its dual role in plant metabolism—acting as both an essential micronutrient and a potential toxicant at higher concentrations [21]-[24]. These findings highlight the need for further research into the biological functions of trace elements within the plant matrix, particularly their role in bioavailability and potential interactions with other dietary components.

In summary, the study underscores the rich phytochemical and mineral composition of *N. sativa* seeds, further substantiating their nutritional and medicinal value. While the presence of key bioactive compounds and essential minerals reinforces their potential therapeutic applications, additional research is necessary to elucidate the mechanisms of action, bioavailability, and potential pharmacological interactions. Future studies focusing on quantitative phytochemical analysis, in vivo bioactivity assessments, and metabolic profiling will provide deeper insights into the full therapeutic potential of *N. sativa*.

3.6. Conclusion

In conclusion, this study affirms the rich phytochemical and mineral profile of *Nigella sativa* seeds sourced from Jos, Nigeria, highlighting their potential nutritional and therapeutic significance. The presence of essential macro- and micro-elements, alongside a broad spectrum of bioactive compounds, reinforces the seeds' value in traditional medicine and as a dietary supplement. While the findings align with previous research from other regions, they also provide localized data supporting the safety and efficacy of *Nigella sativa*. However, to fully harness its medicinal potential, further investigations are warranted to explore the bioavailability, pharmacokinetics, and interactive effects of its constituents within biological systems.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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