Tropical Journal of Phytochemistry & Pharmaceutical Sciences

Available online at https://www.tjpps.org

Original Research Article

Comparative Study of Phytochemical Composition and Antioxidant Properties of *Vigna unguiculata* (Cowpea) Cultivated on Mining and Non-Mining Soils in Edo North

Eseigbe M. Imade ¹* and Usunobun Usunomena²

¹ Department of Biochemistry, Faculty of Basic Medical Sciences, Edo University Iyamho, Edo State, Nigeria ² Department of Biochemistry, Faculty of Basic Medical Sciences, Edo University Iyamho, Edo State, Nigeria.

ABSTRACT

From a nutritional standpoint, Cowpea (Vigna unguiculata) is gaining conspicuous recognition as an important source of proteins, calories, minerals, vitamins and contain significant amounts of polyphenols such as flavonoids, phenolic acids, and lignin, which are considered natural antioxidants. This study evaluated the phytochemical constituents, nutritional composition, and antioxidant properties of cowpea cultivated on mining (Okpella and Ikpeshi) and non-mining (Afuze) soils. Phytochemical screening, proximate composition and antioxidant activity were investigated using standard analytical methods. Phytochemical screening revealed the presence of alkaloids, saponins, phlobatannins, steroids, terpenoids, flavonoids, and phenols in all cowpea extracts. However, Cardiac glycosides with cardenolides (CG3) was absent in all samples. Additionally, Cardiac glycosides with steroidal nucleus, Cardiac glycosides with deoxy sugar (CG1 and CG2) and anthraquinone were exclusively detected in cowpea from the Afuze soil. The highest total phenolic and flavonoid content was recorded for cowpea from the non-mining Afuze site A 34.18 \pm 0.07µg/mg and 27.10 \pm 0.02 µg/mg. Ikpeshi site A demonstrated the highest ABTS, hydroxyl radical scavenging activity and ferric reducing power with 0.89 \pm 0.01 µg/mg, 0.89 \pm 0.04 µg/mg, and 0.80 \pm 0.12 µg/mg respectively while Ikpeshi site B has the highest DPPH activity 0.90 \pm 0.09µg/mg. Conversely, Afuze cowpea had the lowest DPPH, ABTS, OH and ferric reducing power. Non-mining soils consistently yielded cowpea with higher percentages of fat, carbohydrate, nitrogen, dry matter, and protein compared to mining soils. These findings suggest that mining activities negatively impact soil quality leading to decreased nutrient content.

Keywords: Edo North, Mining Impact, Cowpea, Food quality

Received 12 February 2025	Copyright: © 2025 Eseigbe <i>et al.</i> This is an open-access article
Revised 19 February 2025	distributed under the terms of the Creative Commons Attribution
Accepted 27 February 2025	License, which permits unrestricted use, distribution, and reproduction
Published online 01 March 2025	in any medium, provided the original author and source are credited.

Introduction

Mineral extraction in Edo North, Nigeria, presents a notable risk to the region's ecological balance, affecting soil integrity, vegetation, and the well-being of its residents.^{1,2} The process of mining can trigger soil degradation through erosion and compaction, as well as introduce contaminants into both soil and water systems. These changes subsequently impede plant development and diminish agricultural yields. Mining sites release airborne pollutants, including particulate matter, which pose a risk to human respiratory health and can lead to accumulation. Furthermore, heavy metal human-caused contamination of soil, resulting in decreased soil quality, has negative consequences for both public and environmental health. While often challenging to detect due to the soil's inherent capacity to buffer and process pollutants, the cumulative impact can be substantial.³ Soil, the uppermost layer of the Earth's crust, a complex matrix of minerals, organic matter, fluids, gases, and biota, is particularly vulnerable in agricultural settings. The accumulation of heavy metals and related compounds, whether from natural deposits or human-driven activities, contaminates arable land, jeopardizing food safety, availability, and overall environmental sustainability.4

*Corresponding author. Email: eseigbe.mercy@edouniversity.edu.ng

Tel: + 2348062449246

Citation: Eseigbe IM and Usunomena U. Comparative Study of Phytochemical Composition and Antioxidant Properties of *Vigna unguiculata* (Cowpea) Cultivated on Mining and Non-Mining Soils in Edo North. *Trop J Phytochem Pharm. Sci.* 2025; 4(2): 73 – 78 http://www.doi.org/10.26538/tjpps/v4i2.7

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

Cowpea, scientifically known as *Vigna unguiculata*, is a versatile crop, encompassing both agricultural and horticultural applications, and is recognized for its valuable nutritional and therapeutic attributes.⁵ Investigating how mining activities affect the plant's phytochemical makeup, nutritional composition, and antioxidant capacity is critical for evaluating its viability in sustainable farming and its potential for yielding beneficial health compounds. As a member of the *Vigna* genus, which includes various peas and beans, *Vigna unguiculata* is widely recognized as cowpea. The species name, "*unguiculata*," derived from Latin, meaning "possessing a small claw," refers to the distinctive small stalks on its flower petals. All domesticated cowpeas fall under the globally recognized subspecies *V. unguiculata* subsp. *unguiculata*, which is further categorized into four cultivar groups: Unguiculata, *Biflora, Sesquipedalis, and Textilis*.^{5,6}

Cultivated cowpeas are recognized by a variety of common names, including black-eyed pea, southern pea, yard long bean, catjang, and crowder pea.⁸ The term "cowpea" itself was first recorded in American documentation in 1798, likely originating from the plant's historical use as livestock feed.⁹ The black-eyed pea, a name associated with the *unguiculata* cultivar group, is characterized by the prominent dark mark at the seed's hilum. Introduced to the southern United States, early cowpea varieties with densely packed seeds within their pods gave rise to the alternative names, southern pea and crowder pea. *Vigna unguiculata* holds a significant position as a staple legume in sub-Saharan Africa, valued for its ability to enrich soil nitrogen and its utility as animal fodder.¹⁰ Its seeds provide a substantial nutritional profile, containing approximately 24% crude protein, 53% carbohydrates, and 2% fat.¹¹ Furthermore, the leaves and flowers are

edible, and the seeds serve as a rich source of plant proteins, B vitamins, riboflavin, and niacin, all vital for energy metabolism. This research distinguishes itself by examining the specific effects of mining operations within the Edo North region of Nigeria on the nutritional and phytochemical characteristics of cowpeas. While the broader consequences of mining on soil and plant life are understood, this study offers localized data from a potentially less-studied Nigerian area. This focused, regional approach enables a more precise evaluation of the environmental footprint within this specific setting. Therefore, this study aims to determine the extent to which mining influences the phytochemical composition, nutritional content, and antioxidant activities of cowpeas grown in Edo North.

Materials and Methods

Chemical and Reagent

Chemicals such as 1,1-diphenyl-2-picrylhydrazyl (DPPH), 1,10phenanthroline, trichloroacetic acid (TCA), ferric chloride reagent, Dragendorff's reagent, pyridine, sodium nitroprusside reagent, glacial acetic anhydride, and other chemicals/reagents are all products of Evans Medical PLC, Lagos, Nigeria

Sample Collection and Identification

Healthy cowpea seeds (voucher number: 3035612) were obtained from the Department of Crop Science, Leventis Farm, Agenebode, Edo State, Nigeria, in July 2024. The soil collected from each location was loamy sand. Each soil sample was homogenized, crushed, and dried in the dark at room temperature under a fume hood for seven days. One kilogram of each dried soil sample was measured into a planting bag, and six cowpea seeds were planted in each bag. The plants were grown for eight weeks.

Overview of the study area

Edo North, a senatorial district and the second largest administrative division within Edo State, Nigeria, is located in the state's northern region. Geographically, it extends from 6°50'N to 7°30'N latitude and 5°40'E to 6°50'E longitude.¹ This district comprises six local government areas: Akoko-Edo, Etsako Central, Etsako East, Etsako West, Owan East, and Owan West. It shares borders with Kogi and Ondo States to the north, east, and west, and with Uhumwode, Esan West, Esan Central, and Esan North East local government areas to the south. The area under study, situated in northern Edo State, is characterized by sedimentary geological formations, which have contributed to an increase in mining operations. Specifically, Okpella and Ikpeshi, located in the northern part of the district, feature significant geological formations are the source of various extracted materials, including limestone, granite, sandstone, basalt, and chalk.

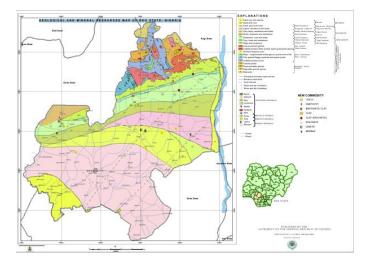


Figure 1: Mineral resources map of Edo State.1

Sample preparation

After eight weeks post-planting, the above-ground portions of the cowpea plants were harvested and thoroughly rinsed with standard tap water. Subsequently, these plant parts underwent a six-week drying period in ambient room temperature, under shade. Following dehydration, the materials were pulverized into a fine powder using a common blending device.

Sample Extraction

A 100g aliquot of each dried plant powder was individually subjected to maceration in 100 mL of a methanol-water solvent mixture (4:1 v/v) within conical flasks for a 24-hour period. Subsequent to this, the resulting mixtures were filtered through Whatman No. 42 filter paper. The filtrate was then subjected to solvent evaporation to yield concentrated plant extracts. These concentrated extracts were meticulously stored in sterilized, airtight containers, appropriately labeled, and maintained at a temperature of 4° C for subsequent utilization

Phytochemical Screening

phytochemical tests for (alkaloids, steroids, flavonoids, phlobatanin, cardio glycosides, phenols, tannins, anthraquinone, saponins and terpenoids) were conducted to identify presence of phytochemicals in the cow pea according to the procedure described by^{12,13}

Determination of the Nutritional Composition

The nutritional composition was evaluated via conventional laboratory techniques, focusing on the quantification of proximate constituents.¹⁴ Following this, available carbohydrate content was determined through a calculation involving the subtraction of the combined percentages of moisture, protein, lipids, ash, and dietary fiber (all expressed on a dry matter basis) from 100%. The energy content of the samples was subsequently calculated utilizing standard energy conversion values: 4 kilocalories per gram (kcal/g) for protein and available carbohydrates, and 9 kcal/g for lipids. This methodology aligns with typical procedures utilized in the nutritional assessment of plant-derived animal feed.

Total Phenolic content

The Folin-Ciocalteu colorimetric assay, following a modified procedure based on.¹⁵, was used to quantify the total phenolic content of the methanolic extracts. A tannic acid standard was prepared by dissolving 0.05g of dry tannic acid in 50mL of distilled water. Serial dilutions of this standard yielded concentrations ranging from 0 to 1.0 μ g/mL. For the assay, 0.5mL of the extract / standard was combined with 10mL of distilled water and 2.5mL of Folin-Ciocalteu reagent. After a two-minute incubation, 7.5mL of 20% sodium carbonate solution was added, and the mixture was diluted to volume with deionized water. After two hours, absorbance was measured at 760nm using a Perkin Elmer Lambda EZ150 spectrophotometer. The results were reported as μ g tannic acid equivalents per mg of sample.

Total Flavonoid Content

The total flavonoid content was measured using a modified protocol derived from. ^{16,17} 0.5 mL of the plant extract, prepared at an appropriate dilution, was mixed with 50 μ L of 10% aluminum chloride, 50 μ L of 1M potassium acetate, and 1.4 mL of distilled water. Following a 30-minute incubation period at room temperature, the absorbance of the resulting solution was determined at 415 nm using a Lambda EZ150 spectrophotometer (Perkin Elmer, USA). A quercetin calibration curve, utilizing concentrations from 0 to 1.0 μ g/mL, was constructed, and the flavonoid content was reported as μ g quercetin equivalents per mg of plant sample

1,1-diphenyl 1-2-picryl-hydrazl (DPPH) Activity of cowpea Extract

To evaluate the antioxidant activity, a DPPH radical scavenging assay was performed, with minor adjustments to the method outlined in.¹⁸ Stock solutions of the samples (4 mg/mL) were diluted in methanol to

achieve final concentrations of 200, 100, 50, 25, and 12.5 $\mu g/ml$. One milliliter of a 0.3 mM DPPH methanolic solution was mixed with one milliliter of each sample dilution. The reaction mixture was incubated at room temperature for 30 minutes, after which the absorbance was determined at 517 nm using a Lambda EZ150 spectrophotometer (Perkin Elmer, USA). Tannic acid was utilized as the reference standard

2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) ABTS radical scavenging Activity of cowpea Extract

The ability to scavenge ABTS radicals was evaluated using a modified procedure based on.¹⁹. The ABTS radical cation was generated by combining 8 mM ABTS solution with 3 mM potassium persulfate and incubating the mixture in darkness at room temperature for 12 hours. A Trolox standard ($1000 \mu g/mL$) was prepared and subjected to serial dilutions to yield concentrations of 12.5, 25, 50, 100, and 200 $\mu g/ml$. To perform the assay, 2.9 mL of the ABTS radical cation working solution was added to 0.1 mL of each Trolox standard concentration, and the reaction was allowed to proceed for 30 minutes. Absorbance measurements were taken at 734 nm for both the standards and samples, using a Perkin Elmer Lambda EZ150 spectrophotometer. A Trolox calibration curve was then constructed by plotting absorbance values against the corresponding concentrations.

Hydroxyl Radical Scavenging Activity of cowpea Extract

The ability of the extracts to protect against Fe^{2+}/H_2O_2 -induced deoxyribose breakdown was evaluated using a method adapted from.²⁰ Stock solutions of each extract were diluted serially in pure methanol to achieve concentrations of 200, 100, 50, 25, and 12.5 µg/ml. A mannitol standard was also prepared and diluted to the same concentration series. In the assay, 1.2 ml of 0.1 M phosphate buffer (pH 7.4) was combined with 7.5 µL of 20 mM hydrogen peroxide, 30 µl of FeCl₃, 45 µl of 1,10-phenanthroline, and 0.5 ml of the extract solution at each concentration. After a 30-minute incubation, absorbance was determined at 532 nm using a Lambda EZ150 spectrophotometer (Perkin Elmer, USA)

Reducing Power Ability of the cowpea Extract

The ferric reducing antioxidant power (FRAP) of the extract was assessed using a modified version of the method described.²¹. Briefly, 0.5 ml of the extract at varying concentrations was combined with 1.25 ml of 0.2 M phosphate buffer (pH 6.6) and 1.25 ml of 0.1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes. The reaction was terminated by the addition of 1.25 ml of 10% trichloroacetic acid. Subsequently, 1.25 ml of the supernatant was mixed with 1 ml of distilled water and 0.25 mL of 0.01% ferric chloride solution. After a 10-minute incubation at room temperature, the absorbance was measured at 700 nm using a spectrophotometer (Perkin Elmer Lambda EZ150) against a suitable blank. Increased absorbance values indicated greater reducing power. Ascorbic acid was used as the standard

Statistical Analysis

Three separate experiments, each with triplicate samples, provided the data for this study. Statistical analysis was conducted using IBM SPSS Statistics (version 20). Group differences were determined through a one-way ANOVA, with subsequent t-tests employed for pairwise comparisons. Significance was established at p < 0.05

Results and Discussion

The phytochemical profiles, proximate composition, and antioxidant activity of cowpea samples collected from mining and non-mining sites are summarized in Tables 1, 2, and 3, respectively. Table 1 indicates that alkaloids, saponins, phlobatannins, steroids, terpenoids, flavonoids, and phenols were present in all cowpea extracts. However, cardiace cardiac glycosides (CG3) were not detected in any sample. Notably, cowpea from the non-mining Afuze site exhibited higher concentrations of phenols, flavonoids, tannins, saponins, and alkaloids. Phlobatannins and steroids showed consistent levels across all sites. Furthermore, steroidal nucleus and deoxy sugar cardiac glycosides (CG1 and CG2), along with anthraquinones, were uniquely identified in cowpea from the Afuze soil. It is well-established that

mining activities can significantly modify soil composition and environmental conditions, which in turn can affect the growth and biochemical characteristics of plants such as cowpea.22 The observed decrease in phenolic compounds, flavonoids, tannins, saponins, and alkaloids in cowpea from mining sites can be attributed to several factors like soil contamination, nutrient imbalance and oxidative stress.²² The influence of mining on plant secondary metabolite production has been shown to be detrimental for example²³ reported that heavy metal stress has been shown to reduce the levels of phenolic compounds, flavonoids, and tannins in various plant species. Anthraquinones are a class of secondary metabolites produced by plants, including cowpea, as a defense mechanism against herbivores and pathogens. These compounds have various biological activities, including insecticidal, antifungal, and antimicrobial properties.24 The presence of anthraquinone in cowpea from non-mining sites but absent in cowpea from mining sites may be attributed to the fact that cowpea plants grown in non-mining soils may naturally produce anthraquinones as a defense against pests and diseases. This is a common strategy used by plants to protect themselves from herbivores and pathogens.24

Table 2 shows non-mining soils consistently yielded cowpea with higher percentages of fat, carbohydrate, nitrogen, dry matter, and protein compared to mining soils. The highest percent of fat, carbohydrate, nitrogen, dry matter and protein are 1.60%, 73.73%, 3.04%, 93.10%, 10.06% respectively. The differences in nutrient composition between cowpea grown on non-mining and mining soils have important implications for human health and nutrition. Cowpea is a valuable source of protein, carbohydrates, and micronutrients.²⁵ Reduced nutrient content in cowpea from mining areas can lead to nutritional deficiencies, particularly in populations that rely heavily on this crop. Cowpea grown on non-mining soils often exhibits higher levels of fat, carbohydrates, nitrogen, dry matter, and protein,22while having lower moisture and ash content compared to those grown on mining soils. This can be attributed to several factors such as hormonal imbalance (Phytohormone Disruption). The release of heavy metals from mining operations can disrupt plant hormonal systems, notably those involving auxins, gibberellins, and cytokinin. These hormonal imbalances interfere with essential plant functions, including growth, development, and nutrient distribution, ultimately leading to diminished growth, altered nutrient usage, and reduced biomass. Lower levels of phenolic and flavonoid compounds observed in cowpea grown on mining soils can be attributed to reduced enzyme activity, impaired substrate availability, and altered gene expression.²⁷ Despite the lower levels of phenolic and flavonoid compounds, cowpea from mining soils often exhibits higher antioxidant activity. This paradoxical effect can be explained by the increased production of other antioxidant compounds, such as ascorbic acid and glutathione, in response to oxidative stress. Additionally, some heavy metal ions themselves may possess antioxidant properties. The altered phytochemical profile of cowpea grown on mining soils has implications for human health. Phenolic compounds and flavonoids have been linked to various health benefits, including antioxidant, anti-inflammatory, and anticancer properties. Reduced levels of these compounds may diminish the potential health benefits of consuming cowpea from mining areas.²¹.

Table 1: Qualitative Phytochemical Screening of Cowpea

 Cultivated in Mining and Non-Mining Soils

Parameters	AF SITE A	AF SITE B	OK SITE A	OK SITE B	IK SITE A	IK SITE B
Alka	+	+	+	+	+	+
Sapo	+	+	+	+	+	+
Phlo	+	+	+	+	+	+
Tan	+	+	+	+	+	+
Anthr	+	+	-	-	-	-
Ster	+	+	+	+	+	+
Ter	+	+	+	+	+	+

Flav	+	+	+	+	+	+
CG1	+	+	-	-	-	-
CG2	+	+	-	-	-	-
CG3	-	-	-	-	-	-
Phenols	+	+	+	+	+	+

IK = Ikpeshi, OK = Okpella, AF = Afuze Ster = Steriods, Terp= Terpernoids, Fla= Flavonroid Anthr = Anthraquinones, Phlo = phlobatannins, Sapo= Saponin, CG1= Cardiac glycosides with steroidal nucleus, CG2 = Cardiac glycosides with deoxy sugar, CG3 = Cardiac glycosides with cardenolides. + = Present, - = Absent,

Table 2: Proximate composition of cow pea cultivated in a mining and non-mining soils

Paramete	r AF SITE A	AF SITE B	OK SITE A	OK SITE B	IK SITE A	IK SITE B
MC (%)	6.07 ± 0.04^{a}	$6.90\pm0.17^{*a}$	18.45 ± 0.06^{ab}	$20.13 \pm 0.02^{*ab}$	20.13 ±0.02*	17.05±0.04*
DM (%)	$93.93\pm3.00^{\rm a}$	$93.10\pm3.00^{\rm a}$	$81.55 \pm 1.00^{\text{b}}$	79.87 ± 0.07^{b}	79.87 ±9.00*a	82.95±2.00*a
Ash (%)	8.32 ± 0.02^{a}	$6.42\pm0.02^{\ast a}$	17.47 ± 0.02^{ab}	$23.65 \pm 0.02^{*ab}$	18.50 ±0.05*	$21.85\pm0.05^*$
O.M (%)	91.68±0.08 ^{ab}	$93.58 \pm 0.08^{*a}$	$82.53\pm0.03^{\ast b}$	$76.35 \pm 0.35^{*ab}$	81.50 ± 0.30^{a}	$78.15 \pm 0.15*$
AIA (%)	$3.02\pm0.10^{\ast b}$	$2.33\pm0.02^{\ast a}$	7.06 ± 0.05^{ab}	$6.13\pm0.04^{*ab}$	$8.10\pm0.02a$	$7.25 \pm 0.04*$
ASA (%)	4.00 ± 2.00^{a}	$3.26\pm0.04^{\rm a}$	6.38 ± 0.07^{b}	$7.00\pm2.00^{\text{b}}$	$6.94 \pm 0.08^{*a}$	$6.08\pm0.04*$
WSA (%)	$4.09\pm0.02^{\rm a}$	$3.46\pm0.12^{\ast a}$	5.00 ± 0.11^{ab}	$5.08\pm0.06^{\ast ab}$	$6.40\pm0.05^{\ast a}$	$5.63 \pm 0.03*$
NC (%)	$3.04\pm0.05^{\ast ab}$	$3.69\pm0.02^{\ast a}$	1.45 ± 0.11^{ab}	1.65 ± 0.00^{ab}	2.33 ± 0.01^{a}	$1.96 \pm 0.05*$
CF (%)	$1.55\pm0.06^{\rm a}$	1.59 ± 0.09^{a}	0.44 ± 0.05^{ab}	$0.60\pm0.02^{\ast ab}$	$0.11\pm0.01^{\ast a}$	$0.24\pm0.04*$
CHO (%)	73.73 ± 0.03^{a}	$73.43\pm0.43^{\rm a}$	58.47 ± 0.47^{ab}	$50.82\pm0.02^{\ast ab}$	55.75 ±0.05*a	$54.22 \pm 0.22*$
Fat (%)	$1.40\pm0.40^{\ast a}$	$1.60\pm1.00^{*b}$	1.19 ± 0.19^{ab}	$1.10\pm0.10^{\ast ab}$	$1.22\pm0.22^{\ast a}$	$1.24\pm0.04*$
CP(%)	$8.90\pm0.00^{\rm a}$	$10.06 \pm 0.04^{*a}$	3.98 ± 0.02^{ab}	$3.70\pm0.12^{\ast ab}$	$4.29\pm0.03*$	$5.90\pm0.03^*$
FE(Kcal)	$343.12\pm0.12^{\rm a}$	$348.36 \pm 0.36^{*a}$	260.51 ± 0.01^{ab}	227.98 ± 7.00**	^{ab} 251.14±0.14*	249.64±0.04*

IK = Ikpeshi, OK = Okpella, AF = Afuze. AIA = Acid insoluble ash, ASA= Acid soluble ash, WSA= water soluble ash. CP. = crude protein, NC = Nitrogen concentration, FE = Food energy, org. matter= organic matter CHO = carbohydrate. Data presented as Mean \pm SD; n = 3; * = p-value less than 0.05 when compared to SITE A; a = p-value less than 0.05 when compared to IK SITE; b = p-value less than 0.05 when compared to OK SITE

Table 3: Antioxidant of cowpea cultivated in a mining and non-mining soil

Total Phenolic content (µgTAE/mg)

Conc.	Samples IK SITE A	IK SITE B	OK SITE A	OK SITE B	AF SITE A	AF SITE B
0.2	7.59 ± 0.03^{a}	$7.11\pm0.01*$	$8.08\pm0.01^{\rm a}$	$7.11 \pm 0.02*$	$13.75 \pm 0.18^{*ab}$	$12.10\pm0.02^{\ast ab}$
0.4	11.41 ±0.03*a	$13.09\pm0.01*$	15.07 ± 0.06^{a}	$13.07 \pm 0.07 *$	$24.29 \pm 0.18^{*ab}$	$21.28\pm0.06^{*ab}$
0.6	14.41 ±0.04*	$16.19\pm0.03*$	$17.90\pm0.10^{\rm a}$	$14.14\pm0.04^{\ast a}$	$28.14\pm0.07^{*ab}$	$24.88\pm0.18^{*ab}$
0.8	17.40 ±0.04*a	$19.98 \pm 0.09*$	$24.03\pm0.05^{\mathrm{a}}$	$19.34 \pm 0.13^{*a}$	$29.36\pm0.09^{*ab}$	$27.24\pm0.08^{*ab}$
1.0	24.15 ± 0.58^{a}	$23.17\pm0.06*$	25.08 ± 0.04^{a}	$21.04 \pm 0.05^{*a}$	$34.18\pm0.07^{*ab}$	$29.79\pm0.02^{*ab}$

Total Flavonoid content (µg quercetin/mg)

Conc. Sample	IK SITE A	IK SITE B	OK SITE A	OK SITE B	AF SITE A	AF SITE B
0.2	$5.09\pm0.02^{*a}$	$5.55\pm0.35*$	$6.16\pm0.03^{\rm a}$	$5.40\pm0.06^*$	11.95 ± 0.06^{ab}	10.18±0.04*ab
0.4	$5.46\pm0.01^{\rm a}$	$6.38\pm0.01*$	$7.42\pm0.02^{\rm a}$	$6.18\pm0.03^{*a}$	16.04 ± 0.05^{ab}	14.11±0.01*ab
0.6	$7.86\pm0.06*$	$8.89\pm0.10^*$	12.37 ± 0.07^{a}	$10.19\pm0.03^{*a}$	19.92 ± 0.05^{ab}	15 .42 ±0.02*ab
0.8	$10.21 \pm 0.01^{*a}$	$12.03\pm0.13^*$	15.20 ± 0.01^{a}	$12.05\pm0.04*$	25.11 ± 0.04^{ab}	$21.21 \pm 0.02^{*ab}$
1.0	12.11 ±0.01*a	$15.16\pm0.05*$	$18.08\pm0.02^{\rm a}$	$12.49\pm0.05^{\ast a}$	27.10 ± 0.02^{ab}	23.10±0.01*ab

DPPH (µgTAE/mg)

Conc. Sample 12.5 25 50 100	$\begin{array}{c} \text{IK SITE A} \\ 0.84 \pm 0.05^a \\ 0.82 \pm 0.02^a \\ 0.81 \pm 0.03^{*a} \\ 0.79 \pm 0.01^* \end{array}$	$\begin{array}{c} \text{IK SITE B} \\ 0.90 {\pm} 0.09 {*} \\ 0.88 {\pm} 0.04 {*} \\ 0.84 {\pm} 0.00 {*} \\ 0.77 {\pm} 0.02 {*}^a \end{array}$	$\begin{array}{c} OK \ SITE \ A \\ 0.85 \pm 0.10^{*a} \\ 0.82 \pm 0.04^{*a} \\ 0.81 {\pm}.03^{*b} \\ 0.76 \pm 0.01^{a} \end{array}$	$0.89 \pm 0.15^{*a}$	$\begin{array}{l} AF\ SITE\ A\\ 0.75\ \pm\ 0.03^{ab}\\ 0.73\ \pm\ 0.10^{ab}\\ 0.71\ \pm\ 0.04^{ab}\\ 0.60\ \pm\ 0.02^{a} \end{array}$	$\begin{array}{l} AF \ SITE \ B \\ 0.78 \pm 0.12^{ab} \\ 0.76 \pm 0.04^{*a} \\ 0.77 \pm 0.01^{*ab} \\ 0.73 \pm 0.01^{*ab} \end{array}$
200	$0.75\pm0.01^{\rm a}$	$0.77\pm0.00*$	$0.71\pm0.01^{\rm a}$	$0.75\pm0.02^{\ast a}$	0.59 ± 0.01^{ab}	$0.62\pm0.00^{*ab}$
			ABTS (µgTE	/mg)		
Conc. Sampl	e IK SITE A	IK SITE	B OK SITE	A OK SITE I	B AF SITE A	AF SITE B
12.5	$0.89 \pm 0.01^{\circ}$	ka 0.87±0.02	$3* 0.76 \pm 0.12$	2^{ab} 0.82 ± 0.02	*b 0.60 ± 0.20 ^{ab}	$0.71 \pm 0.00a$
25	0.82 ± 0.04 *	ka 0.85*±0.0	$0.03* 0.75 \pm 0.20$	$0.81 \pm 0.10^{\circ}$	*a 0.59 \pm 0.01 ^{ab}	$0.68 \pm 0.00^{*ab}$
50	0.81 ± 0.12^{a}	0.84±0.1	$7^{*a} = 0.72 \pm 0.02$	3^{a} 0.81 ± 0.02	*a 0.57 \pm 0.02 ^b	$0.64 \pm 0.04^{*ab}$
100	$0.72 \pm 0.10^{\circ}$	0.82 ± 0.0	0.69 ± 0.02	2^{a} 0.77 ± 0.01	*a 0.55 ± 0.01 ^{ab}	0.62±0.02*ab
200	0.70 ± 0.20	a 0.80±0.1	0^{*a} 0.64 ± 0.0	0.1^{ab} 0.72 ± 0.02	*a 0.50 ± 0.20 *b	$0.50\pm0.20^{\rm a}$
		Hydi	roxyl (OH) (µ	gTE/mg)		
Conc. Sample	IK SITE A	IK SITE B	OK SITE A	OK SITE B	AF SITE A	AF SITE B
12.5	$0.89\pm0.04^{\rm a}$	0.87 0.02*	$0.82 \pm 0.01^{*a}$	$0.87 \pm 0.03^{*a}$	0.70 ± 0.20^{ab}	$0.73\pm0.03^{*b}$
25	$0.86 \pm 0.03^{*a}$	$0.84 \pm 0.00*$	0.81 ± 0.00^{a}	$0.85 \pm 0.02^{*a}$	0.68 ± 0.01^{ab}	$0.71 \pm 0.01^{*ab}$
50	0.84 ± 0.02^{a}	0.82±0.11*a	0.81 ± 0.00^{a}	$0.82 \pm 0.10^{*b}$	0.67 ± 0.03^{ab}	$0.68\pm0.02^{*ab}$
100	0.82 ±0.010*	0.81±0.02*a	$0.79\pm0.03^{*ab}$	$0.80 \pm 0.10^{*a}$	0.62 ± 0.02^{ab}	0.64 ± 0.04^{ab}
200	$0.80 \pm 0.17*$	$0.78\ 0.01^{*a}$	$0.75 \pm 0.02^{*a}$	0.78 ± 0.02^{ab}	0.60 ± 0.20^{a}	$0.61\pm0.00^{*ab}$
			FRAP (µgTE	/mg)		
Conc. Sample	E IK SITE A	IK SITE B	OK SITE A	OK SITE B	AF SITE A	AF SITE B
12.5	$0.80\pm0.12^{\rm a}$	$0.84 \pm 0.04^{*a}$	$0.78 \pm 0.02*$	$^{\rm b}$ $0.88 \pm 0.04^{\circ}$	* 0.59 ± 0.01^{ab}	0.64 ± 0.03^{ab}
25	$0.84\pm0.10^{*a}$	$0.83\pm0.02^{\ast a}$	$0.75\pm0.04^{\rm a}$	$0.85 \pm 0.04*$	0.57 ± 0.02^{ab}	$0.62\pm0.12^{*ab}$
50	$0.81\pm0.03^{*a}$	$0.82\pm0.11*$	$0.73\pm0.03^{\rm a}$	$0.83 \pm 0.03*$	0.56 ± 0.01^{ab}	$0.61 \pm 0.10^{*ab}$
100	$0.77\pm0.02*$	$0.81\pm0.00^{\rm a}$	$0.72\pm0.02*$	ab 0.80 ± 0.10^{b}		0.60 ± 0.20^{ab}
200	$0.72\pm0.01^{\rm a}$	$0.80\pm0.10^*$	$0.67 \pm 0.00^{*a}$	^b $0.76 \pm 0.04^{*}$	0.51 ± 0.01^{ab}	$0.64 \pm 0.04^{*ab}$

Data presented as Mean \pm SD; n = 3; * = p-value less than 0.05 when compared to SITE A; a = p-value less than 0.05 when compared to IK SITE; b = p-value less than 0.05 when compared to OK SITE. IK = Ikpeshi, OK = Okpella, AF = Afuze.

Conclusion

The results obtained demonstrate the adverse effects of mining on both agricultural yields and the quality of food produced in the region. The compromised nutritional profile and modified phytochemical constituents of cowpea grown in mining soils may have significant repercussions for human health, potentially contributing to nutrient inadequacies and a reduced capacity to combat chronic diseases. To alleviate these effects, the implementation of environmentally responsible mining practices is essential to minimize soil deterioration and pollution. Moreover, the creation of crop cultivars that exhibit increased resistance to heavy metal exposure and other environmental pressures is vital. Future investigations should focus on exploring the specific mechanisms driving these changes and on formulating methods to improve the nutritional content and functional characteristics of cowpea cultivated in mining areas.

Conflict of Interest

The authors declare no conflict of interest.

Author's Declaration

The authors hereby declare that the work presented in this article is original. Any liability for claims relating to this article will be borne by us.

Acknowledgement

This research was made possible by the generous support of the Tertiary Education Trust Fund (TETFUND) through the 2024 Institution-Based Research (IBR) Grant. We extend our sincere gratitude to the laboratory staff of the Department of Biochemistry, Edo University Iyamho, Nigeria, for their invaluable assistance, and to Mr. Kenneth O. for his support.

References

- Idris1 GN, Asuen GO, Ogundele OJ. Environmental Impact on Surface and Ground Water Pollution from Mining Activities in Ikpeshi, Edo State, Nigeria. Int. J. Geosci. 2016; 5:749-75
- Okhakhu PA. Rural Development and Environmental Protection in Nigeria. Dev. Countr. Stud. 2016; 6:1-6
- Ming⁻ate FLM, Mohamed MY. Impact of stone quarrying on the environment and the livelihood of communities in Mandera County. Kenya. J. Sci Res. Rep. 2016; 10:1-9.
- Osuocha KU, Akubugwo EI, Chinyere GC, Ugbogu AE. Seasonal impact on phytoaccumulation potentials of selected edible vegetables grown in Ishiagu quarry mining effluent discharge soils. Afr. J. Environ. Sci. Technol. 2016; 10:34-43.
- Du SK, Jiang H, Yu X, Jane JL. Physicochemical and functional properties of whole legume flour. LWT Food Sci. Technol. 2014; 55:308–313

- 6. Awika JM, Duodu KG. Bioactive polyphenols and peptides in cowpea (*Vigna unguiculata*) and their health promoting properties: A review. J. Funct. Foods 2017;38: 686–697.
- Onyelucheya CM, Nwabanne TJ, Onyelucheya OE, Onuoha OE. Dilute acid hydrolysis of cowpea hulls: A kinetic study. Int. J. Adv. Sci. Eng. Inf. Technol. 2016; 6:451–455.
- Li G, Zhu F. Physicochemical properties of quinoa flour as affected by starch interactions. Food Chem. 2017; 221:1560–1568
- 9. Cui Y, Wang Y, Li Y. Wang Y, Liu Y. Phytochemical composition and antioxidant activity of different parts of *Moringa oleifera*. J. Food Sci and Technol. 2018;55(11):4523-4532.
- Stagnari F, Maggio A, Galieni A, Pisante M. Multiple benefits of legumes for agriculture sustainability: an overview. Chem Bio. Technol. Agric. 2017; 4(1):2
- 11. Food and Agriculture Organization (FAO). Grassland species index. *Vigna unguiculata L*, 1986-2012
- 12. Ijoma KI, Ajiwe VIE, Odinma SC. The organic extracts from the leaves of *Ficus thonningii Blume, Jatropha tanjorensis* J.L Ellis and Saroja and *Justicia carnea* Lindley as potential nutraceutical antioxidants and functional foods. Trends Phytochem. Res. 2023; 7(1): 76-85
- 13. Siyuan S, Tong L, Rui HL. Corn phytochemicals and their health benefits. Food Sci. Hum. Wellness 2018; 7:185–195
- AOAC. Off. Methods Anal. 11th ed., Association of Official Analytical Chemists, USA, 1995, p. 1015
- Singleton VL, Rossi JA. Colorimetry of total phenolics with phosphomolybdic phosphotungstic acid reagents. Am. J. Enol. Vitic 2015; 16:144–158.
- Ghasemzadeh A, Jahangir M. Antioxidant activities and phenolic compounds of selected medicinal plants. J. Med. Plants Res. 2016;10(15): 173-182.
- Cui Y, Wang Y, Li Y. Wang Y, Liu Y. Phytochemical composition and antioxidant activity of different parts of *Moringa oleifera*. J. Food Sci.Technol. 2018;55(11):4523-4532.

- Indrianingsih AW, Wulanjati MP, Windarsih A, Bhattacharjya DK, Suzuki T, Katayama T. Antioxidant and α-glucosidase inhibitor activities of natural compounds isolated from *Quercus gilva Blume* leaves Biocatal. Agric. Biotechnol. 2021;12(5):213-218
- Re R, Pellegrini N, Proteggente A, Pannala, A, Yang M, Riceevans. Antioxidant activity applying an improved ABTS radical decolorization assay. Free Radic. Biol. Med. 2019;26;1231-1237.
- Yu W, Zhao Y, Shu B. The radical scavenging activities of *Radix puerariae* isoflavonoids: A chemiluminescence study. Food Chem. 2016; 86:525-529.
- Benzie, IF, Strain JJ. The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power" the FRAP assay. Anal. Biochem. 2016; 239:70-76
- Yunlong H, Zhifeng Y, Xiangling F, Weixiong Z, Jinrong L, Feng Z Influence of Mining and Vegetation Restoration on Soil Properties in the Eastern Margin of the Qinghai-Tibet Plateau. Int. J. Environ. Res. Public Health. 2020;174-288
- Michael OA, Jiřina S, Pavel T. The fate of secondary metabolites in plants growing on Cd-, As-, and Pb-contaminated soils—a comprehensive review. Environ. Sci. Pollut. Res. 2023; 30:11378– 11398
- Marco M, Antonio E. Fungal Bioactive Anthraquinones and Analogues Toxins. Food Chem 2020; 12:714
- 25. Felix DD, Alphonsus KB. Evaluation of Protein and Micronutrient Levels in Edible
- Cowpea (Vigna Unguiculata L. Walp.) Leaves and Seeds. Frontiers: In Sustain. Food Syst. 2019;3-17
- Ze L, Takeshi H, Nobutoshi Y, Toshiro I. The Roles of Plant Hormones and Their Interactions with Regulatory Genes in Determining Meristem Activity. Int. J. Mol. Sci.2019;20; 4065