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Research Article

Evaluation of the Nutritional Quality, Bioactive Compounds, and Antioxidant Activity of *Moringa oleifera* Leaf Extract

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ABSTRACT

This study was conducted to evaluate the proximate and mineral compositions, phytochemical content, and antioxidant activity of *Moringa oleifera* leaf extract to determine its nutritional quality and medicinal attributes. The proximate, mineral, and phytochemical contents were evaluated using standard methods. The result of proximate compositions revealed that *Moringa oleifera* leaves contain ash (10.12±0.3), moisture (7.10±0.1), protein (19.05±0.2), fat (2.03±0.01), fiber (13.5±0.02), and carbohydrates (48.20±1.02). This showed that *Moringa oleifera* leaves are rich in proteins and fiber and have low moisture and high ash contents. The results of the mineral analysis indicated that the leaves of *Moringa oleifera* are rich in minerals, the concentration of the mineral elements in µg/g are iron (3.15±0.01), potassium (5.10±0.03), calcium (6.24±0.01), zinc (1.14±0.001), magnesium (4.62±0.02), and copper (2.02±0.03). The ethanolic extract of *Moringa oleifera* leaves was analyzed for phytochemicals, the results indicated the presence of alkaloids (3.31±0.03 g/100g), flavonoids (5.01±0.1 g/100g), glycosides (0.25±0.001 g/100g), anthraquinones (3.03±0.05 g/100g), terpenoids (5.45±0.01 g/100g), tannins (2.09±0.02 g/100g) and phenols (13.89±0.1 g/100g). The DPPH and ABTS radical scavenging assay showed that *Moringa oleifera* leaf extract can scavenge free radicals. However, the highest inhibition was 90.98 % for DPPH and 95.14% for ABTS, all at 100 mg/ml. The high antioxidant activity observed could be attributed to the high concentration of bioactive compounds, especially phenols and flavonoids. Based on these findings, it could be inferred that *Moringa oleifera* leaves could be used as a source of nutrients and bioactive compounds that can be used in the management and treatment of chronic diseases.

Keywords: Antioxidant Activity; Minerals; *Moringa oleifera*; Proximate; Phytochemicals

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INTRODUCTION

Chronic non-communicable diseases are the leading cause of morbidity and mortality worldwide with an estimated reported cases of 523 million suffering from CVD and estimated 41 million death representing 71% of all deaths. Out of the estimated 41 million death, 17.9 million are related to cardiovascular diseases, 9.3 million are cancer related, 4.1 million related to respiratory diseases, 1.5 million from diabetes while the remaining 8.2 million are attributed to other forms of non-communicable diseases (Hacker *et al*., 2020; Taheri, 2024; Odunyemi *et al*., 2024). Most of the deaths are premature and occurred in low and middle income countries. The World Bank predicted that, by the year 2030 the non-communicable diseases related deaths would surpass infectious diseases related death in Africa (Marquez and Farrington, 2023). Nigeria is among the countries with biggest burden of non-communicable diseases, in 2016

premature deaths from non-communicable diseases represents 29% of the total deaths. Moreover, the greater part of health related expenditure is attributed to NCDs, perhaps this has contributed to the high rate of poverty among NCDs patients (Ezenwaka *et al*., 2024).

In addition to genetic factors, factors responsible for the development of chronic non-communicable diseases include environmental pollution, consumption of junk and processed foods and life style among others (Allender *et al*., 2019; Khonje *et al*., 2019; Reyes-Olavarría *et al*., 2020). These factors contributes to the generation of free radicals and reactive oxygen species in the body leading to oxidative stress (Engwa *et al*., 2022). It is a well-known fact that oxidative stress is strongly correlated to the incidence of chronic diseases such as cancer, hypertension, obesity, diabetes mellitus, coronary heart diseases, chronic bronchitis and so on (Jomoba *et al*., 2023; Sharifi-Rad *et al*., 2020). Epidemiological study has indicated that air pollution is closely related to respiratory diseases such as chronic obstructive pulmonary diseases and has contributed to the global diseases burden mainly due to exposure to $PM_{2.5}$ (Duan *et al*., 2020). On a global scale, the World Health Organization reported that about 7 million people die each year due to exposure to air pollutants out of which 3.3 million are due to cardiovascular diseases. This major and expanding health risk is linked to the urbanization of the world population and being exposed to contaminated air (Kim *et al*., 2020). Pollutants cause oxidative stress and local inflammation when they reach the lungs, which then causes systemic inflammation. This inflammation, in-turn, causes endothelial dysfunction, thrombosis, and increased atherosclerosis (Arujo, 2011). Consumption of junk and high-caloric food has been linked to various chronic illnesses, including diabetes mellitus, hypertension, cardiovascular diseases, metabolic syndrome, stroke, neurological diseases and obesity, among others, all as a result of the generation of reactive oxygen species (Singh and Vellapandian, 2023; Murugesan and Mahendraprabu, 2024). Free radicals and reactive oxygen species are known to damage proteins, DNA and membrane lipids leading to pathological consequences (Jomova *et al*., 2023).

Synthetic antioxidants and modern drugs that are used to treat chronic conditions are characterized by a certain level of toxicity (Stoia and Oancea, 2022). On the other hand natural antioxidants derived from food and other natural sources are effective and safer than synthetic ones (Garcia *et al*., 2013). Plants are known to exhibit various biological activities such as antioxidant, anti-inflammatory, antihypertensive, antibacterial and hypolipidemic properties among others. These biological properties are mostly attributed to the presence of bioactive active compounds including flavonoids, short peptides, vitamins, terpenoids and phenolic compounds (Josa *et al*., 2024). Most of these compounds act as antioxidants, having the ability to scavenge free radicals and reactive oxygen species, thereby preventing oxidative stress which in turn contribute to overall health (Samitaya *et al*., 2021; Santos, 2021). *Moringa oleifera* is a plant that originated from India and now present in various parts of the world including Africa (Popoola and Obembe, 2013; Soto *et al*., 2025). *Moringa oleifera* is known to possess various medicinal properties which could be due to the presence of bioactive compounds (Calmday-Ombo and Eluehike, 2025; Villegas-Vazquez *et al*., 2025). Different parts of *Moringa oleifera* such leaves, bark, fruits, roots, flower and seeds are consumed for therapeutic purpose, in fact the leaves and flower are consumed as food (Rahman *et al*., 2021). This study intends to evaluate nutritional quality and antioxidant activity of *Moringa oleifera* leaf extract.

MATERIALS AND METHODS

Sample Collection and Preparation

A sample of *Moringa oleifera* leaves was obtained from the cultivated farm of Aminu Kano College of Islamic and Legal Studies in Kano metropolis and was authenticated at the Department of Biology, Aliko Dangote University of Science and Technology Wudil. The leaves were washed and cleaned with distilled water and dried at room temperature for five days. The dried sample was processed into a powder using a mortar and pestle and then stored in an air-tight container.

Determination of Mineral Elements

The determination of mineral elements such as iron (Fe), potassium (K), calcium (Ca), zinc (Zn), magnesium (Mg), and copper (Cu) employed the method of AOAC (2005). The technique is based on the observation that when biological materials are exposed to high temperatures (600°C for five hours), their organic components are entirely burned, leaving behind the inorganic components that might be used to identify and measure mineral elements. Two grams (2g) of the powdered sample of *Moringa oleifera* leaves was placed in a crucible and transferred to a muffle furnace and was heated up to a temperature of 600°C for 5 hours to obtain a white ash. The ash was transferred to a desiccator and was allowed to cool at a room temperature. The ash was digested using 5 mL of 10% hydrochloric acid and the volume was made to 50 mL with distilled water. Atomic absorption

spectrophotometer was used to analyze the concentration of the mineral elements.

Atomic Absorption Spectrophotometry (AAS)

This technique is based on the principle that atoms of lower energy level absorbs energy in form of ultraviolet or visible light and get excited to a higher energy level. The wavelength of light that was transmitted is measured by the detector and then compared with the wavelength of light passing through the sample. When the processor integrates the wavelength shift, the peak of energy absorption at a specific wavelength is displayed (Farrukh, 2012).

Atomatic Absorption Spectrophotometry Analysis

To carry out the analysis, standard solutions of the mineral elements in question were prepared in accordance with the standard protocols. Both the sample, standard and blank were aspirated into the flame. Thereafter, the elemental ions were atomized and absorbed a radiation of characteristic wavelength from a hollow cathode. The absorbance is proportional to the amount of the element in the sample.

Determination of the Proximate Parameters

Moisture contents were determined by calculating weight loss due to evaporation of moisture in a hot air oven and weight loss after drying to a constant weight. The ash content was determined by igniting 5 g of the powdered sample of *Moringa oleifera* leaves in a muffle furnace, which is based on the idea that all organic matter has the potential to burn and release carbon (IV) oxide and water. Crude fat was determined using a Soxhlet extractor, the sample was placed in a Soxhlet apparatus and extracted using petroleum ether. The extract was collected in a flask and evaporated, leaving behind the crude fat, which was weighed. Crude protein is primarily composed of protein and other nitrogenous non-protein molecules like amides and ammonium compounds. This was calculated from the samples' total nitrogen by multiplying the nitrogen content of foods by 6.25. The insoluble and combustible organic residue that is left after the sample has been subjected to light petroleum ether treatments, boiling in diluted sulfuric acid and sodium hydroxide, and washing with boiling water, alcohol, and petroleum ether is the crude fiber. Crude fiber was calculated as percentage of weight loss due to combustion at 550^0C in a muffle furnace. The carbohydrate content was calculate by difference as follows;

100 - (Moisture + Ash + crude fat + crude Protein + crude fiber) (Mikail et al., 2023).

Determination of Phytochemical Contents and antioxidant Activity

Sample Extraction

For phytochemicals and antioxidant activity determination, 20g of the powdered sample of *Moringa oleifera* leaves was dissolved in 100 mL of 80% ethanol and was allowed to soak overnight. The solution was filtered using whatman No. 1 filter paper and was stored in a clean container.

Phytochemical Screening

The leaves extract of *Moringa oleifera* was screened for the presence of phytochemical compounds. This is with a view to qualitatively and quantitatively determined the presence of alkaloids, flavonoids, anthraquinones, glycosides, terpenoids, tannins and phenols. This was carried out in accordance with the methods described by Velvan *et al*. (2015) with little modification.

Determination of antioxidant Activity

The (DPPH) radical-scavenging activity was determined as described by Selamassakul *et al*. (2016). 200 µL of *Moringa oleifera* sample extract was added to 2 mL of DPPH• (0.1 mM) which is prepared by adding 1mM DPPH in 9mL of 80% ethanol. The mixture was shaken and incubated in the dark for 30 min. The absorbance was monitored at 517 nm using spectrophotometer. Distilled water was used as a blank. The scavenging effect was calculated using the following equation:

$$\%\text{DPPH radical scavenging Activity} = \frac{\text{ABlank} - (\text{ASample} + \text{AControl})}{\text{ABlank}} X\ 100$$

Where A_{blank} = Absorbance of of 1mL dstilled water + 2mL of 0.1 mM ethanolic DPPH solution

A_{Sample} = Absorbance of 1 mL aqueous sample + 2 mL of 0.1mM ethanolic DPPH solution

$A_{Control}$ = Absorance of 1 mL aqueous sample + 2 mL of 80% Ethanol

3.4.6.2 2, 2^1-azino-bis 3ethylbenzthiazoline-6-sulfonic (ABTS•+) Radical Scavenging Assay

The ABTS scavenging effect of *Moringa oleifera* was carried out in accordance with the methods described by Selamassakul *et al*. (2016). The reagent (ABTS•+) was made by adding 5 mL of 7 mM ABTS•+ to 7 mM potassium sulfate (K_2SO_4) at a ratio of 2:1. The solution was incubated in the dark room for 12 to 16 hours to obtain a maximum radical generation. Thereafter, the solution was diluted with distilled water to obtain an absorbance of 0.700 ± 0.030 at 734 nm before use. One milliliter (1mL) of the ABTS•+ reagent was added to 10 µL of *Moringa oleifera* extract and stored for 6 min in the dark. The absorbance was measured at 734 nm using distilled water as a blank. The antioxidant

activity of the sample was calculated using the same equation as that applied in the DPPH• test.

RESULTS

Proximate and Mineral Composition

The proximate composition of *Moringa oleifera* leaves are presented in table 1. The result indicated that *Moringa oleifera* leaves have high carbohydrate, protein and fiber contents, and are low in fat contents. Table 2 presents the concentration of mineral elements of *Moringa oleifera* leaves. The result indicated that the leaves of *Moringa oleifera* have high amount of calcium, potassium and magnesium and low amount of zinc and copper.

Qualitative and Quantitative Phytochemical Contents

Table 3 presents the qualitative phytochemical contents of ethanolic leaves extract of *Moringa oleifera*. Based on the results, the leaves of *Moringa oleifera* contained alkaloids, flavonoids, anthraquinones, glycosides, terpenoids, tannins and phenols. As presented in table 4, compounds of highest concentration are phenolic compounds, terpenoids and flavonoids.

Antioxidant Activity

The antioxidant activities of ethanolic leaves extract of *Moringa oleifera* were determined by DPPH and ABTS radical scavenging assays and the results are presented in figure 1. This was determined at various concentrations (40 mg/mL, 60 mg/mL, 80 mg/mL and 100 mg/mL) respectively. The % DPPH and ABTS antioxidant activities. The results indicated that the ethanolic leave extract of *Moringa oleifera* possess antioxidant activity which increase as the concentration of extract increased. However, the ABTS assay showed higher radical scavenging activity when compared to the DPPH assay.

Table 1: Proximate Composition of *Moringa oleifera* Leaves

Parameters (%)	Composition
Ash	10.12±0.3
Moisture	7.10±0.1
Protein	19.05±0.2
Fats	2.03±0.01
Fiber	13.5±0.02
Carbohydrates	48.20±1.02

Table 2: Mineral composition of *Moringa oleifera* Leaves

Mineral Elements	Concentration (µg/g)
Iron	3.15±0.01
Potassium	5.01±0.03
Calcium	6.24±0.01
Zinc	1.14±0.001
Magnesium	4.62±0.02
Copper	2.02±0.03

Table 3: Qualitative Phytochemical Contents of Ethanolic Leaves Extract of *Moringa oleifera*

Compound	Concentration g/100g
Alkaloids	++
Flavonoids	+++
Anthraquinones	++
Glycoside	+
Terpenoids	+++
Tannins	++
Phenols	+++++

Table 4: Quantitative Phytochemical Contents of Ethanolic Leaves Extract of *Moringa oleifera*

Compound	Concentration g/100g	
Alkaloids	3.31±0.03	
Flavonoids	5.01±0.1	
Anthraquinones	3.03±0.05	
Glycoside	0.25±0.001	
Terpenoids	5.45±0.01	
Tannins		2.09±0.02
Phenols		13.89±0.1

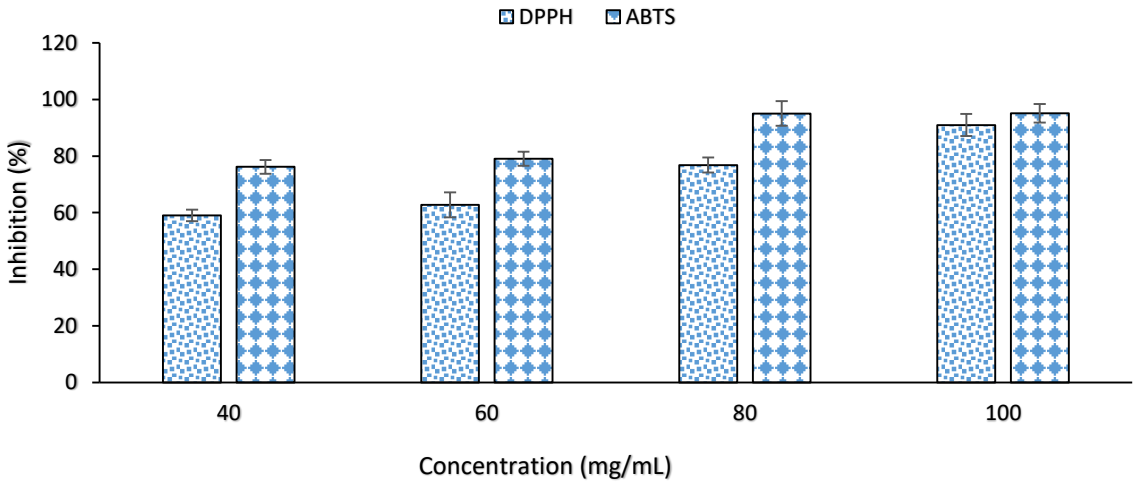

Figure 1: DPPH and ABTS radical Scavenging Activity of Ethanolic Leaves Extract of *Moringa oleifera*

DISCUSSION

Proximate and Mineral Compositions

The nutritional value of vegetables such as *Moringa oleifera* leaves can be assessed through their proximate and mineral contents (Ambo *et al*., 2023). In this study, the proximate and mineral contents of *Moringa oleifera* were determined with a view to ascertaining their nutritional quality. The proximate composition of *Moringa oleifera* leaves are presented in table 1. The result indicated that the leaves of *Moringa oleifera* are rich in carbohydrates (48.20±1.02%), protein (19.05±0.2%) and fiber (13.05±0.02%). The results also indicated that *Moringa oleifera* leaves have low moisture (7.10±010%) and high ash (10.12±0.3%) contents. The low moisture content is an indication that the leaf powder of *Moringa oleifera* can be stored for a long time without deterioration, since lower moisture contents increase the shelf life of foods (Gichau *et al*., 2020; Long *et al*., 2022). On the other hand, the high ash contents implied that *Moringa oleifera* leaves could be rich in minerals, this is because the concentration of mineral elements of foods is determined by ash contents, and the higher the ash contents the higher the minerals (Mlonka-Mędrala *et al*., 2020). The protein content of *Moringa oleifera* leaves determined in this study is higher than that of *Telfairia occidentalis* (3.55%), *Solanum nigrum* (9.22%) reported by Ezema *et al*. (2024), *Piper guineense* (18.61%) reported by Aremu *et al*. (2024) and comparable to that of *Senecio biafrae* (19.54%) reported by Ezema *et al*. (2024). The carbohydrate contents of *Moringa oleifera* leaves reported in this study are lower than that of *Telfairia occidentalis* (51.63%), Solanum nigrum (55.90%) reported by Ezema *et al*. (2024), and higher than that of Amaranthus (29.32%), *Corchorus olitorius* (8.38%), *Senecio biafrae* (48.47%) reported by Ezeme *et al*. (2024), *Piper guineense* (43.62%), *Spinach* (26.13%) and *Gongrenema latifolium* (40.38%) reported by Aremu *et al*. (2024). In terms of fiber contents, the value observed in this study are higher that of *Piper guineense* (11.09%), Spinach (8.52%) reported by Aremu *et al*. (2024), *Senecio biafrae* (8.48%), *Telfairia occidentalis* (10.46%), *Amaranthus* (8.45%), *Solanum nigrum* (8.78%) reported by Ezema *et al*. (2024), *Piper guineense* (11.09%) and Spinach (8.85%) reported by Aremu *et al*. (2024). The lipid contents (2.03%) of *Moringa oleifera* leaves used in this study is lower than those of various leafy vegetables (Aremu *et al*., 2024; Ezema *et al*., 2024; Jibrin *et al*., 2024).

Plant proteins have been shown to be inferior when compared with animal proteins, however, plant proteins have been reported to have advantages over animal proteins. For instance, consumption of plant proteins have been shown to decreased the risk of developing chronic diseases such as cardiovascular diseases. On the other hand, proteins from animals, especially those from red meat have been linked to the development of cardiovascular diseases (Huang *et al*., 2020). Moreover, consumption of plants that are rich in proteins will provide other bioactive compounds such as vitamins and flavonoids that could improve overall health (Ullah *et al*., 2020; Shen *et al*., 2022). Therefore, *Moringa oleifera* could be used as a source of protein most especially in vegetarian diet. Plant foods most especially vegetables have been shown to provide carbohydrate of higher quality to the global food supply and are widely promoted for their positive impact on human health (Schulz and Slavin, 2021; Drewnowski *et al*., 2022). Carbohydrates are important source of energy and

metabolic intermediates required for proper functioning of cells (Sultana and McClure, 2023). Belorkar *et al*. (2016) reported that the carbohydrates present in vegetables includes fructo-oligosaccharides, due to their prebiotic property, fructo-oligosaccharides are considered as functional foods (Kherade *et al*., 2021). Therefore, *Moringa oleifera* could be used as a source of prebiotic. Dietary fiber is important in human nutrition, this is because fiber has been shown to have blood cholesterol lowering effect, reduced risk of diabetes mellitus and cardiovascular diseases (Alhassan *et al*., 2015). Based on the findings of this study, it could be asserted that *Moringa oleifera* leaves have proximate composition that could enhance the wellbeing of individuals and could also be used to improve food security. In terms of mineral contents, it could be asserted that *Moringa oleifera* leaves could be used as a source of minerals (table 2). The predominant mineral elements of *Moringa oleifera* leaves are calcium, potassium, magnesium and iron. Mineral elements have a wide range of functions in cell. They are required as cofactors for various enzymatic reactions and are important for other cellular activities. Magnesium have the ability to binds to organic substances such as proteins and nucleic acids, it plays roles in enzyme activation, membrane function and signaling, calcium is involved in bone health, zinc is involved in DNA and RNA synthesis, acts as antioxidant, enhance immunity and regulates the formation of hormones. Magnesium, copper, zinc and iron are required for proper functioning of the immune system (Ciosek *et al*., 2021; When *et al*., 2022). Therefore, *Moringa oleifera* leaves can be used as a source of mineral elements and could therefore be considered as supplement.

Phytochemical Compositions

Phytochemicals are naturally occurring compounds that are found in plants. They have a wide range of biological activities that have positive impact on human health (Melini and Ruzzi, 2025). The phytochemical contents of the leaves of *Moringa oleifera* are presented in table 3. Based on the results, phytochemical compounds of high concentration in the leaves of *Moringa oleifera* includes phenols (13.89±0.1 g/100g), terpenoids (5.45±0.0 g/100g) and flavonoids (5.01±0.1 g/100g). Alkaloids include various compounds having diverse medicinal properties including anti-inflammatory, anti-fungal, anti-tumor, antibacterial and neuroprotective activities among others (Li *et al*., 2025). Alkaloids includes quinine, melorfomine, ajamaline, camptothecin, sanguinarine and so on. Alkaloids possess biological activities due to the presence of nitrogenous heterocyclic rings in their structure that allow them to easily interact with biological system and confer biological effect (Debnath *et al*., 2018). The biological activities of flavonoids have been widely reported (Dwivedi *et al*., 2017). Phenolics and flavonoids have been shown to possess anti-aging, antioxidant, anti-inflammatory, antidiabetic antihypertensive, antimicrobial and hypolipidemic attributes (Brahmachari *et al*., 2011; Yang *et al*., 2025; Zheng *et al*., 2025; Liu *et al*., 2025; Alharbi *et a*l., 2025). Flavonoids includes quercetin, salicylic acid, chlorogenic acid, coumaric acid, rutin, Kaempferol, apigenin, luteolin, myricetin and many more (Dabeek *et al*., 2019; Roy *et al*., 2022). The presence of hydroxyl groups are key to biological activities of phenols and flavonoids. This is because the hydroxyl groups have the ability to donate electrons to free radicals thereby preventing oxidative damage to cells and tissues (Dias *et al*., 2021). Moreover, phenols and flavonoids act as meal chelators, having the ability to bind metal ions and prevent oxidative damage (Ghasemzadeh and Ghasemzadeh, 2011). Anthraquinones are types of phenolic compounds with different biological activities such as antioxidant and antibacterial properties (Qun *et al*., 2023). Glycosides are naturally occurring steroids compounds found in plant. Due to their ability to inhibit Na^+/K^+, glycosides act as cardiotonic agents and are commonly used in the treatment of cardiac diseases Škubník *et al*., 2021). As with other phytochemical compounds, terpenoids exhibited various medicinal properties. They have been reported to have antioxidant, antimicrobial, anticancer, anti-inflammatory and antiviral properties and are therefore used as natural source of drugs Surowiak *et al*., 2021).

Antioxidant Activity

The antioxidant properties of the leaves of *Moringa oleifera* as determine by DPPH and BTS radical scavenging assays are presented in figure 1. The results showed that *Moringa oleifera* leaves extract has the ability to scavenge free radicals. However, the scavenging activity increase with increase in concentration of the extract (figure 1). However, the ABTS radical scavenging assay showed higher antioxidant properties at all concentrations. This could be attributed to the fact that ABTS is soluble in both polar and nonpolar solvents and is therefore able to determine the antioxidant capacity of both hydrophobic and hydrophilic compounds (Ácsová *et al*., 2019). Based on these findings, it could be asserted that, the antioxidant activity of *Moringa oleifera* leaves extract is attributed to the presence of its phytochemical compounds such as flavonoids, phenols and terpenoids presented in table 2. The antioxidant activity of *Moringa oleifera* determined

in this study is higher than that of Fenugreek Seed reported by Ibrahim *et al*. (2020).

CONCLUSION

Findings of this study revealed that *Moringa oleifera* leaves can be used as good source of micro and macronutrients and could therefore be used to address food insecurity and malnutrition. Moreover, the leaves can be used as a source of bioactive compounds having numerous biological activities including antioxidant activity. The leaves extract have the ability to inhibit radicals generated by DPPH and ABTS at all concentrations, however the highest inhibitory activities for DPPH (90.98%) and ABTS (95.14%) was observed at 100 mg/mL. The high and antioxidant activity of *Moringa oleifera* leaves could be due to its phytochemical content.

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Competing Interest

The authors have declared that no competing interest exist

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