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Influence of Location on the Phytochemical, Nutritional and Mineral Composition of the Leaves of *Moringa oleifera* (LAM) Grown in Ekiti State

Sunday Arowosegbe[1, *], Mary Kehinde Olanipekun[2], Odunayo James Olowoyeye[3]

Authors'Affiliation
[1,2,3]Department of Plant Science and Biotechnology, Faculty of Science, Ekiti State University, PMB 5363, Ado-Ekiti, Nigeria.

***Corresponding Author:**
Sunday Arowosegbe
Department of Plant Science and Biotechnology, Faculty of Science, Ekiti State University, PMB 5363, Ado-Ekiti, Nigeria.

E-mail:
Sunday.arowosegbe@eksu.edu.ng

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Abstract

Moringa oleifera is a versatile plant commonly used as medicine and as vegetable for meeting the nutritional needs of many communities in Nigeria and all over the world. This study was carried out to determine the effects of location on the phytochemical, nutritional and mineral composition of M. oleifera leaves in Ekiti State, Nigeria. Seeds of Moringa were planted in four locations made up of two towns each from the tropical rain forest (Efon-Alaaye and Ikere-Ekiti) and the derived Savannah (Omuo-Ekiti and Otun-Ekiti) areas of the State. Leaf samples were harvested after 90 days and analyzed for their phytochemical, proximate and mineral composition, using standard methods. The results showed the presence of some major phytochemicals, proximate and Mineral elements in different proportion across locations. The leaves of M. oleifera grown in Ikere Ekiti consistently produced the highest quantities of alkaloids (0.18 mg/g), saponins (0.16 mg/g), phenols (38.70 mgGAE/g) and tannins (14.91 mgTAE/g). Interestingly, leaves of the plant harvested from Efon Alaaye also had high quantity of these phytochemicals. Significantly highest proximate composition in the leaves was found to be10.33 % Ashin Efon-Alaaye, 30.26% protein in Ikere Ekiti, and 11.05% Crude fiber in Omuo Ekiti. However, no significant differences ($p \leq 0.05$) was recorded in Carbohydrate and Fat contents irrespective of location. Moreover, leaves harvested from Ikere Ekiti and Efon Alaaye had the highest proximate content. Appreciable quantity of Na and Mg were also discovered in the leaves from OmuoEkiti.The best combination of phytochemicals, proximate and minerals were found in the plant samples grown in Ikere Ekiti and Efon Alaaye, while the least was in samples from Otun Ekiti. Farmers are to therefore, carefully consider location and soil factors in the cultivation of M. oleifera.

INTRODUCTUION

Medicinal plants are of great importance to the health of an individual and communities at large and their medicinal importance lie in the chemical substances called bioactive ingredients or

phytochemicals that produce a definite physiological action on human body (Okwu, 2004; Olanipekun *et al*., 2020). They are known to contain substances which could be useful in the treatment of ailments directly or used as bases to produce drugs. Many plants known to alleviate symptoms of illnesses have been screened to have substances of medicinal importance, *Moringa oleifera* being one of such plants.

M. oleifera plant is 5-15 m in height with soft brittle stems (Ijarotimi *et al*., 2013), having a diameter of about 30 cm when fully mature. The leaves are compound, pinnate double and of small round or oval in shape. The plant is considered as one of the world's most useful trees. It was noted by Arowosegbe (2019) that *M. oleifera* plays a significant role in the livelihood of many communities in the developing countries of the world as food and for medicinal purposes. Khalafalla *et al*. (2010) also reported that the plant can be used for industrial purposes apart from being used for food and medicine. *M. oleifera* is a common and popular vegetable in different parts of Nigeria. Keay (1989) reported that the plant is known by different names across the three major ethnic groups in the country; 'Zogallagandi' (by the Hausas), 'Ewe igbale' (by the Yorubas) and 'Okwe Oyibo' (by the Igbos).

The high cost of acquiring modern medicine and their inadequate supplies in most health care facilities, the side effects associated with their use and the belief that plants hold cure for many disease conditions have led to a reawakening of interest in the use of plants and plant products in recent years among the people of West Africa (Minja, 1999). *M. oleifera* have been reported for the treatment of ailments such as stomach disorder, fever, diarrhea, skin infection, as well as antitumor, antipyretic, antiepileptic, anti-inflammatory, antiulcer, antihypertensive and with antioxidant properties (Bukar *et al*., 2010). All these properties had been attributed to the presence of phytochemicals in this plant.

Phytochemicals are compounds that occur naturally in plant parts such as flowers, buds, leaves, fruits, roots and stems/ stem barks. They act as defensive mechanism for the plant against outbreak of diseases and many external attacks and also contribute to the colour, flavor and smell of the plants. Phytochemicals play many ecological and physiological roles and they are widely distributed as plant constituents or secondary metabolites. They include alkaloids, saponins, tannins, steroids, phenols and flavonoids (Okwu, 2004). Interestingly, the abundant nutrients present in the leaves of *M. oleifera* leaves makes it basic and important factors for the selection of the plant for nutritive value, systematic classification and plant improvement programs (Nisar *et al*., 2009). Since some medicinal plant species are used as food in addition to their medicinal benefits, evaluating their nutritional significance can help in understanding the worth of these plant species (Pandey *et al*., 2006).

Most importantly, the totality of any plant species (both physical and chemical properties) is the product of its genetic make-up and the influence of environment on the plant. According to Heldt (2005), the environment could affect the synthesis of secondary metabolites and nutritional compositions irrespective of the medicinal plant species. Incidentally, it has been reported that climatic factors such as rainfall, temperature, light, relative humidity and stage of maturity have much influence on the growth and development of plants and these could cause variation in distribution of the Phytochemicals and nutritional contents in them (Gardener *et al*., 2010). This study was therefore carried out to evaluate the phytochemical and nutritional and proximate composition of the leaves of *M. oleifera* grown in different locations of Ekiti State; with a view of knowing the effects of these locations on the studied parameters.

MATERIALS AND METHODS

Description of the study area

The study was carried out using *M. oleifera* planted in four different locations in Ekiti State, Nigeria. The locations are made up of two towns each from the tropical rain forest (Efon-Alaaye and Ikere-Ekiti) and the derived Savannah (Omuo-Ekiti and Otun-Ekiti) areas of the State. Efon-Alaaye lies within latitude 7.70°N and longitude of 4.96°E of the Greenwich meridian, with an average annual temperature of 24.6°C and precipitation of 1361 mm. Ikere-Ekiti lies within latitude 7.50°N and longitude 5.21°E of the Greenwich meridian, with an average annual temperature of 25.4°C and

precipitation of 1355 mm. Otun-Ekiti lies within latitude 7.98°N and longitude 5.11°E, with average annual temperature of 24.7°C and precipitation of 1292 mm. Omuo-Ekiti lies within latitude 7.76°N and longitude 5.72°E, with average annual temperature of 24.3°C and precipitation of 1296 mm. (Adelade and Fagbemi, 2017).

Land Preparation, Soil Analysis and Seed Sowing

A total land area of 9 m x 18 m was used for the study in each location. Soil samples were collected randomly from the upper layer to a depth of 1 to 15 cm in each of the study locations and transported to the laboratory to determine the physical and chemical composition of the soil samples. The land was cleared manually using machete. Heaps were made after packing the refuse with a spacing of 1m x1m between rows. Seeds used for this study were collected from mature and dry pods of a *M. oleifera* plant. Plant samples were also collected and taken to the herbarium of the Department of Plant Science and Biotechnology, Ekiti State University, Ado-Ekiti, Nigeria for authentication. Two seeds were planted per hole at a depth of 2-3 cm at a spacing of 100 cmx100 cm. The seedlings were reduced to one plant per stand after emergence. Scheduled weeding was done throughout the period of planting across the locations.

Collection and Preparation of Plant Samples for Phytochemical, Proximate and mineral Analyses

Fresh leaves of the *M. oleifera* plant were harvested across the locations 90 days after planting, washed under running tap, drained and air-dried for two weeks to avoid volatilization. The air-dried samples were ground into powder using an electric blender before subjecting them to phytochemical, proximate and mineral analyses.

PHYTOCHEMICAL ANALYSIS

Qualitative Phytochemical analysis of sample

The leaf samples were analyzed for the presence of Phytochemicals as follows.

Test for Tannins

Using the method of Trease and Evans (2005), 0.5g each of the dried samples was boiled in 20 mL of distilled water in a test tube and then filtered. A few drops of 0.1% ferric chloride was added and observed for brownish green or blue colouration, indicating the presence of tannins.

Test for Saponins

Using the method of Harbone (1994), 2 g each of the powdered samples was boiled in 20 mL of distilled water in a water bath and filtered. 10 mL of the filtrate was mixed with 5 mL of distilled water and shaken vigorously for a stable persistent froth. The frothing was mixed with 3 drops of olive oil, shaken vigorously and then observed for the formation of emulation.

Test for Alkaloids

The method of Sofowora (2006) was adopted. Each of the crude powdered samples (5 g) was defatted with 5% ethyl ether for 15 min and extracted for 20 min with 5 mL aqueous HCL on a boiling water bath. The resultant mixture was centrifuged for 10 min at 3000 rpm. One millimeter (1 mL) of the filtrate was treated with few drops of Mayer's reagent and another 1 mL with Dragendoff's reagents. Creamish/brown/red/ orange/ precipitate was observed indicating the presence of Alkaloids.

Test for Terpenoids

Each extract (5 mL) was mixed with 2 mL of chloroform and 3 mL of concentrated H_2SO_4 was carefully added to form a layer (Trease and Evans, 2005). A reddish brown colouration of the interface was formed to show positive results for the presence of terpenoids.

Test for Flavonoids

Using the method of Mayuri (2012), 2 mL each of the sample extracts was placed in each test tube, heated with 10 mL of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4 mL of

the filtrate was shaken with 1 mL of dilute ammonia solution. A yellow colouration was observed indicating a positive test for flavonoids.

Test for Phlobatannins
Deposition of a red precipitation when an aqueous extract of each samples was boiled with 1% aqueous hydrochloric acid was taken as an evidence for the presence of phlobatannins (Sofowora, 2006).

Test for Phenols
Using the method of Tiwari *et al*. (2011), each of the powdered samples (10 mg) was treated with 2 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenol.

Test for Anthraquinones
Using the method of Herbone (1994), Samples was extracted with ether and the filtrate aqueous ammonia is added. Red, pink or violet colour produced indicated the presence of anthraquinones

Quantitative Phytochemical Analysis of Sample

***Determination of Alkaloid contents*:** Using Harbone (1993) method, 5 g each of the powdered sample was weighed into a 250 mL beaker and 200 mL of 10% acetic ethanol was added, covered and allowed to stands for 4 min. This was filtered and the extract was concentrated in a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitation was collected and washed with dilute ammonium hydroxide and then filtered.
The residue is the alkaloid, which was dried and weighed
% Alkaloid= W3-W2/W1 x 100
Where,
W1=Weight of sample.
W2= Weight of dish.
W3=Weight of dish + filtrate after drying.

Determination of Flavonoid contents
The amount of flavonoids in the extract of each of the *M. oleifera* samples was determined by using the aluminum colorimetric assay method (Arowosegbe *et al.,* 2012). A volume of 0.5 mL of the sample solution was added to 0.5 mL of 2% $AlCl_3$ ethanol solution. After 1 h at room temperature, the absorbance was measured at 420 nm using UV spectrophotometer. Extracts samples were evacuated at a final concentration of 0.1 mg/mL. Total flavonoids were calculated as mg/g of quercetin standard curve using the following calibration:

Y = 0.0255x, R2 = 0.9812, where x was the absorbance and Y as the quercetin equivalent.

Determination of Saponin contents
The spectrophotometric method of Brunner (1994) was used for saponins analysis. Each of the finely ground powdered samples (1 g) was weighed into a 250 mL beaker and 100 mL of isobutyl alcohol was added; the mixture was shaken on a UDY shaker for 5 h to ensure uniform mixing. Thereafter, the mixture was filtered into a 100 mL beaker and 200 mL of 40% saturated solution of magnesium carbonate was added. The mixture obtained with saturated $MgCo_3$ was again filtered to obtain a clear colourless solution. A volume of 1 mL of the colourless solution was pipetted into 50 mL volumetric flask and 2 mL of 5% $FeCl_3$ solution was added and made up to mark with distilled water. This was allowed to stand for 30 min for blood red colour to develop. A volume of 0-10 ppm standard saponin solutions were prepared from saponin stock solution. The standard solutions were treated similarly with 2 mL of 5% $FeCl_3$ solution as done for 1 mL sample above. The absorbance of the sample as well as standard saponin solutions were read after colour development in a Jenway V6300 Spectrophotometer at a wavelength of 380 nm.

Saponin content = $\frac{\text{Absorbance of sample x gradient factor x dilution factor}}{\text{Weight of sample x 10,000}}$

Determination of Tannin contents

Using Swain (1979) method, 0.20 g of the sample was measured into a 50 mL beaker, 20 mL of 50% methanol was added and covered with parafilm and placed in a water bath at 77-78^{0}C for 1 h. It was shaken thoroughly to ensure a uniform mixing. The extract was then filtered into a 100 mL volumetric flask and 20 mL water was added. Afterwards, 2.5 mL Folin- Denis reagent and 10 mL of 17% Na_2CO_3 were added and mixed properly. The mixture was made up to mark with water mixed well and allows standing for 20 min.

The bluish- green colour will develop at the end of the range, 0-10 ppm were treated similarly as 1 mL sample above. The absorbance of the tannic acid standard solutions as well as samples was read after colour development on a spectrophotometer at a wave length of 760 nm. Results were expressed as milligrams of Tannic Acid Equivalents (mg TAE/g) using the calibrated curve from the equation: Y = 0.0593x - 0.0485; R2 = 0.9826, where x was the absorbance and Y tannic acid equivalent.

Determination of Phenolic contents

Using the method of Devmurari (2010), 2 g each of the powdered samples was extracted with 20 mL of acetone with 0.5% formic acid for 2 min and was filtered. A volume of 2 mL of the extract was mixed with 0.5 mL Folin-Ciocalteu reagent for 15 Sec and allow standing at 40^{0}C for 30 min. The absorbance was measured at 765 nm and expressed as milligrams of Gallic Acid equivalents (mg GAE/g).

Proximate analysis

The standard methods of Udo and Oguwele (1986) and the Association of Official Analytical Chemists (AOAC, 2012) were used for the determination of moisture, ash, crude fiber, carbohydrate, proteins and fat contents in the leaf samples of the *M. oleifera.*

Mineral Composition

The mineral contents were determined on aliquots of the solution of the dried leaf powdered sample by UV/visible and atomic absorption spectrophotometer (AOAC, 2012). A quantity of 1 g each of the powdered samples was weighed into a pyrex beaker and 10 mL of the concentrated HNO_3 was added and allowed to soak for 30 min. Then, 3 mL of 60% perchloric acid was added. The sample was placed on hot plate and heated at 350^{o}C until frothing stopped and HNO_3 almost evaporated. Then, watch glass was placed on the beaker and heating continues until the sample turn light straw in colour. This was then removed from the hot plate and cooled. The watch plate was then rinsed into the sample and the sample filtered into 100 mL volumetric flask and made up to the mark with distilled water. This was analyzed using flame photometer for sodium and potassium and AAS for other minerals.

DATA ANALYSIS AND RESULTS

The results obtained from phytochemical, mineral and proximate analyses were subjected to Analysis of Variance (ANOVA). The mean values obtained from samples across the locations were separated using Tukey's Multiple Range Tests at $P < 0.05$.

Table 1: Chemical and Physical Properties of Soil Samples at Different Planting Locations of the study areas in Ekiti State.

Soil Parameters	Location			
	Forest Area		Savannah Area	
	Efon Alaaye	Ikere Ekiti	Otun Ekiti	Omuo Ekiti
Chemical properties				
pH	5.40±0.02^c	6.18±0.04^a	5.92±0.02^b	6.12±0.05^a

Organic Carbon (%)	0.99±0.01^{c}	2.07±0.03^{a}	0.74±0.02^{d}	1.41±0.07^{b}
Organic Matter (%)	1.71±0.11^{c}	3.57±0.02^{a}	1.36±0.04^{d}	2.43±0.15^{b}
Available Phosphorus (mg/kg)	28.75±1.03^{c}	123.90±1.57^{a}	63.25±0.57^{b}	5.60±0.86^{d}
*CEC (Cmol/kg)	4.47±0.52^{d}	22.28±0.36^{a}	6.16±0.11^{c}	10.19±0.18^{b}
E.A (Cmol/kg)	2.40±0.05^{a}	0.16±0.02^{c}	0.12±0.01^{cd}	0.24±0.01^{b}
Elect. Cond. (µ S)	118.50±1.11^{c}	174.50±1.03^{a}	85.00±0.53^{d}	132.10±1.07^{b}
Calcium (Cmol/kg)	2.29±0.10^{d}	17.07±0.18^{a}	4.24±0.08^{c}	7.35±0.13^{b}
Magnesium (Cmol/kg)	0.59±0.01^{c}	1.91±0.03^{a}	0.56±0.01^{cd}	0.92±0.02^{b}
Sodium (Cmol/kg)	0.34±0.02^{a}	0.33±0.01^{a}	0.24±0.01^{b}	0.23±0.01^{b}
Potassium (Cmol/kg)	1.14±0.02^{c}	2.86±0.04^{a}	0.98±0.02^{d}	1.45±0.01^{b}
Nitrogen (%)	0.25±0.02^{c}	0.48±0.03^{a}	0.22±0.01^{cd}	0.34±0.01^{b}
Physical Properties				
Sand (%)	81.00±0.14^{b}	80.00±0.10^{b}	87.00±0.21^{a}	88.00±0.16^{a}
Silt (%)	14.60±0.23^{a}	12.60±0.11^{b}	6.60±0.12^{c}	6.60±0.10^{c}
Clay (%)	4.40±0.05^{d}	7.40±0.01^{a}	6.40±0.03^{b}	5.40±0.02^{c}
Textural class	Sandy loam	Sandy loam	Sandy loam	Sandy loam

*CEC = Cation Exchange Capacity, E.A =Electron Affinity, Elect. Cond =Electrical conductivity.

Qualitative Phytochemical Composition of the Leaves of *Moringa oleifera* in different locations of the Study Area

The results of the qualitative phytochemical composition of the leaves of *Moringa oleifera* across locations revealed the presence of Alkaloids, Tannins Phenols, Saponins and Flavonoids, while Terpenoids, Anthraquinons and Phlobatannins were absent in samples from all the locations (Table 2).

Table 2: Qualitative Phytochemical Composition of the Leaves of *Moringa oleifera* from different locationsin the Study Areas of Ekiti State

Phytochemicals		**Location**		
	Forest Area		**Savannah Area**	
	EfonAlaaye	**Ikere Ekiti**	**Otun Ekiti**	**Omuo Ekiti**
Alkaloids	+	+	+	+
Tannins	+	+	+	+
Phenols	+	+	+	+
Saponins	+	+	+	+
Flavonoids	+	+	+	+
Terpenoids	-	-	-	-
Anthraquinons	-	-	-	-
Phlobatannins	-	-	-	-

Key: +=present, - = absent

Phytochemical Composition of the Leaves of *Moringa oleifera* in different locations of the Study Area

The results of the quantitative phytochemical analysis of the leaves of *M. oleifera* from different locations in Ekiti State are shown in Table 3. As revealed in the results, the level of these phytochemicals in the plant varied from location to location. For example, the leaves of *M. oleifera*

plant grown in Ikere Ekiti in the Forest area of the state consistently produced the highest quantities of alkaloids (0.18 mg/g), saponins (0.16 mg/g), phenols (38.70 mgGAE/g) and tannins (14.91 mgTAE/g). Interestingly, the leaves of the plant harvested from EfonAlaaye (another town in the forest area of the state), also had high quantity of these phytochemicals with a value 0.16 mg/g, 0.14 mg/g, 32.60 GAE/g and 12.43 mgTAE/g respectively. However, the least saponins (0.13 mg/g), phennols (30.41 mgGAE/g) and tannins (8.29 mgTAE/g) was found in the leaves of *M. oleifera* fromOtun Ekiti, a town in the savannah area. The leaves of plants harvested from Omuo Ekiti (Forest area) produced the smallest quantity alkaloids (0.12 mg/g) and the highest quantity of flavonoids (18.25 mgQE/g).

Table 3: Quantitative Phytochemical composition of *Moringa oleifera* from towns in the different study areas in Ekiti State

Area	Towns	Alkaloids (mg/g)	Saponins (mg/g)	Phenols (mgGAE/g)	Tannins (mgTAE/g)	Flavonoids (mgQE/g)
Forest	Efon	0.16±0.00^{b}	0.14±0.00^{b}	32.60±0.12^{b}	12.43±0.02^{b}	12.45±0.03^{d}
	Ikere	0.18±0.00^{a}	0.16±0.00^{a}	38.70±0.06^{a}	14.91±0.01^{a}	16.40±0.23^{b}
Savannah	Otun	0.16±0.00^{b}	0.13±0.00^{c}	30.41±0.01^{c}	8.29±0.05^{d}	14.24±0.02^{c}
	Omuo	0.12±0.00^{c}	0.16±0.00^{a}	32.73±0.07^{b}	11.29±0.05^{c}	18.25±0.10^{a}

Values followed by the same alphabet in the same column are not significantly ($p \leq 0.05$) different from each other using Tukey's Multiple Range Test.

The proximate composition of *Moringa oleifera*

The results of the analysis of proximate compositions of *M. oleifera* leaves grown in the towns from both the rain forest and savannah areas are presented in Table 4. There seemed to be no significant differences ($p \leq 0.05$) in the carbohydrate and fat contents of the Moringa leaves irrespective of the location of planting. However, the least moisture content was recorded in leaves harvested from EfonAlaye (9.60%), while the highest moisture content (12.56%) was in the leaf samples from Ikere Ekiti. Leaves of Moringa from Efon Alaaye also had the highest Ash content (10.33%), though not significantly different from that of Otun Ekiti (10.22%). The least Ash content was however found in the leaves harvested from Omuo Ekiti. Interestingly, the highest protein content was discovered in the leaves of Moringa plants grown in Ikere Ekiti (30.26%), followed by Omuo Ekiti (29.55%), while the least was in the samples from Otun Ekiti (26.96%).The significantly highest crude fiber content was found in the leaves harvested in Omuo Ekiti (11.05%), followed by Otun Ekiti (10.78%), both towns being in the Savannah area of Ekiti State. However, the least crude fiber was in the leaves of Moringa plants from Ikere Ekiti (10.46%) and Efon Alaaye (10.32%).

Table 4: Proximate composition of *Moringa oleifera* leaves from towns in the different study areas in Ekiti State

Area	Towns	Moisture (%)	Fat (%)	Ash (%)	Protein (%)	Crude Fiber (%)	Carbo-hydrate (%)
Forest	Efon	9.60±0.05^{c}	1.63±0.81^{a}	10.33±0.03^{a}	28.94±0.01^{c}	10.32±0.06^{c}	38.35±0.16^{a}
	Ikere	12.56±0.06^{a}	2.45±0.04^{a}	9.50±0.06^{b}	30.26±0.08^{a}	10.46±0.02^{c}	37.17±0.09^{a}
Savannah	Otun	10.16±0.03^{b}	2.62±0.03^{a}	10.22±0.01^{a}	26.69±0.05^{d}	10.78±0.02^{b}	37.13±0.14^{a}
	Omuo	10.15±0.03^{b}	2.78±0.01^{a}	9.27±0.01^{c}	29.55±0.04^{b}	11.05±0.01^{a}	37.19±0.11^{a}

Values followed by the same alphabet in the same column are not significantly (p≤0.05) different from each other using Tukey's Multiple Range Test.

The mineral composition of *Moringa oleifera*

The results of the mineral analysis of the leaves of *M. oleifera* harvested from different locations in the study area (Table 5) indicated that the mineral availability varied from one location to another. Moringa leaves harvested from Ikere Ekiti (Forest Area) had the significantly highest Na (123.00mg/100g), K (1435mg/100g) and Ca (5190mg/100g) compositions; while the highest contents of Mn (44.36mg/100g) Mg (685mg/100g), Fe (23.25), Zn (5.18mg/100g) and Cu (1.86mg/100g) were discovered in the leaves of Moringa harvested from Efon Alaaye (Forest Area). Appreciable quantity of Na (122.50mg/100g) and Mg (685mg/100g) was also discovered in the leaves of Moringa harvested from Omuo Ekiti. Interestingly, the lowest estimated quantities of these mineral elements were found in the leaf samples from Otun Ekiti.

Table 5: Mineral composition (mg/100g) of *Moringa oleifera* leaves from different towns in the study areas of Ekiti State.

Area	Towns	Na	K	Mn	Mg	Fe	Ca	Zn	Cu
Forest	Efon Alaaye	117.00 ±1.15^{b}	1218.00 ±1.15^{b}	44.36 ±0.44^{a}	692.33 ±1.45^{a}	23.25 ±0.14^{a}	4427.50 ±1.44^{c}	5.18 ±0.05^{a}	1.86 ±1.71^{a}
	Ikere Ekiti	123.00 ±0.58^{a}	1435.00 ±2.89^{a}	22.60 ±0.12^{b}	607.33 ±1.45^{b}	18.55 ±0.03^{b}	5190.00 ±5.77^{a}	3.37 ±0.04^{c}	1.76 ±1.61^{ab}
Savannah	Otun Ekiti	103.00 ±0.58^{c}	874.75 ±2.74^{d}	23.40 ±0.06^{b}	515.00 ±2.89^{c}	8.83 ±0.04^{d}	4445.00 ±2.89^{c}	3.47 ±.017^{c}	1.32 ±1.27^{c}
	Omuo Ekiti	122.50 ±1.44^{a}	1170.00 ±5.77^{c}	22.50 ±0.06^{b}	685.00 ±2.89^{a}	11.35 ±0.20^{c}	4925.00 ±14.43^{b}	4.17 ±0.03^{b}	1.64 ±1.54^{b}

Values followed by the same alphabet in the same column are not significantly (p≤0.05) different from each other using Tukey's Multiple Range Test.

DISCUSSIONS

The presence of alkaloids, flavonoids, tannins, phenols, saponins in the leaves of *Moringa oleifera* in all the locations where it was grown (Table. 2), justifies the medicinal importance of the plant. These phytochemicals are biologically active and they are required for the curative and preventive purposes as reported by Sofowora (1993) and Okwu (2005)., Hence, their ethnomedicinal uses (Sofowora, 1993; Okwu, 2005; Ogundele *et al*., 2017). Ali (2012) specifically reported that alkaloids have antitumor, antiviral, antihypertensive, antidepressant, anti-inflammatory and antimicrobial activities. The presence of alkaloids in the leaves of *Moringa oleifera* attested to their uses in the management of various diseases. Edeoga and Enata (2011) also reported that alkaloids are powerful pain reliever, having an antipyretic action, a stimulating effect and can act as tropical anesthetic in ophthalmology. Similarly, the flavonoids found in the *M. oleifera* leaves harvested across the locations could be responsible for its use as anti-bacterial, anti-inflammatory, anti-oxidant, anti-allergies, antiviral, anti-mutagenic and vasodilatory properties as reported by Sofowora (1993). Saponins found in the leaves of *M. oleifera* across locations could be an evidence of having the properties of binding cholesterol and hemolytic activity in aqueous solutions (Sodipo*et al.,* 2000). Saponins are also known to impact the immune system and possess cholesterol lowering potential that has been demonstrated in animals and human trials (Guclu-Ustundag and Mazza, 2007). Additionally, phenols have been reported to be a good anti-aging, anti-carcinogen and good cardiovascular protecting agent (Yadav and Agarwala, 2011). Similarly, tannins could be effective in curing hemorrhage as well as restriction of bare swellings. When applied internally, tannins affect the walls of the stomach and contract or squeeze the membrane thereby, restricting secretions from the cells. Hence, appropriate use of herbs possessing tannins are widely used as mouth washes, eyes washes, snuff and even as virginal douches and also in treating rectal disorder (Elvin-Lewis *et al.,* 1977).

As discovered in this study, the quantity of these phytochemicals in the *M. oleifera* leaf samples (Table. 3) seemed to be affected by location as reflected in the highest content of alkaloids, saponins, phenols and tannins in the leaf samples collected from Ikere Ekiti in the rain forest area of the state. This could be attributed partly to the prevailing environmental condition in this location. For example, a look at the chemical and physical properties of the soil samples (Table. 1) indicated that the soil from Ikere Ekiti had the best combinations of these soil attributes. The implication of this is that, the good soil conditions in Ikere Ekiti might have been responsible for more phytochemical contents of the leaves of Moringa grown there. Arowosegbe (2016) had earlier reported that improved soil condition did not only affect the growth and development of plant (*Hibiscus sabdariffa)*, it could also lead to an increase in the phytochemical contents of the plant.

The results of proximate analyses of the leaf samples of Moringa revealed the presence of major nutrients in the *M. oleifera* leaf samples from all the locations. However, the contents were found to vary with location. This finding corroborates the earlier assertions (Sabale *et al.*, 2008; Fowoyo and Oladoja, 2015), that the growing condition, season, age of the plant as well as the soil where a plant is grown could affect the nutritional composition of the plant. Brady and Well (2001) also reported that the dietary constituent of the leaves of M. *oleifera* is as a result of the essential amino acid which could be accumulated as a result of the influence of locations and stages of development of the plant.

As revealed in this study, the protein values obtained in the leaf samples of *M. oleifera* in Ekiti state varied significantly from location to location. However, they are higher than the ones reported for the same plant by Lamidi *et al.,* (2017) in Ara (27.64%), Igbon (25.27%), Ejigbo (23.93%) and Oko (24.33%) which are towns in Osun and Oyo States of Nigeria respectively. More so, the highest proportion of proteins in the sample from Ikere Ekiti (30.26%) could be surmised to be as a result of higher capacity of the soil to exchange cations/anion (CEC) or simply the soil's ability to release its nutrient. This is likely to be enhanced by availability of more water in the soil in this town being a rain forest area (Table 1). However, the *M. oleifera* leaf is relatively rich in protein irrespective of location since they have more than 12% of the calorific values from protein according to Pearson and Cuthbertson (1996). Thus, the appreciable proportions of proteins (in all locations) subjected the plant to be a good source of supplementary protein for man and livestock. Relative amount of carbohydrate was also discovered in the leaves of the *M. oleifera* in all locations. Hence, this establishes the fact that the leaves can be ranked as being carbohydrate- rich. Sufficiency of carbohydrate is necessary for optimum functioning of the brain, hearth, nervous, digestive and immune systems. Moisture content was found to have significantly increased in Ikere Ekiti (12.56%) when compare to the least proportion in Efon-Alaaye (9.60%), thus, the result of the moisture contents across locations is favorably in agreement with the findings of Fugile (2001). This study also revealed the presence of crude fiber in all the *M. oleifera* leaf samples. Crude fiber had been reported to cleanse digestive tract by removing potential carcinogens from the body and hence prevents the absorption of excess cholesterol. In addition, crude fiber adds bulks to food and reduces the intake of excess starchy food which is the characteristics of the diet of the poor and local people. This therefore, guards against adverse metabolic conditions such as hypertension and diabetes in man (Sabale *et al.,* 2008). Ash and fat contents were of bearable limit across locations. The percentage composition of ash and fat (across location) is in line with the research reports of Mulaudzi *et al.* (2012). The low level of fat in the investigated plant justifies that this plant is good for health.

The mineral composition of the leaves of *M. oleifera* across location (Table 5.) reveals high concentrations of Calcium (Ca), Potassium (k), Sodium (Na), Manganese (Mn), Magnesium (Mg), Iron (Fe), Copper (Cu) and Zinc (Zn). Thus, the presence of abundant mineral (in highest proportion) in Ikere Ekiti and Efon Alaaye Ekiti could be as a result of the locations and as influenced by the CEC of the soil (Table 1). Interestingly, the mineral compositions across locations are in agreement with the values reported by Reedy and Bhatt (2001) as he claimed that variations in mineral nutrients could results from differences in location, soil type, age of cultivation and climatic changes.

CONCLUSION

The results of this study revealed the presence of appreciable quantity of alkaloids, flavonoids, phenols, saponins, tannins and different nutrients in different proportions in *Moringa oleifera* which is medicinally and nutritionally important. However, the quantities of all these were found to vary from location to location. The best combinations of phytochemicals, proximate and mineral contents were found in the leaf sample of *M. oleifera* grown and harvested from Ikere Ekiti and Efon Alaaye (in the Rain Forest Area of Ekiti State). This could be traced to the fact that the properties of soil (best in Ikere Ekiti) among others, varied from one location to the other.

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