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Variations in the mineral composition and heavy metals content of *Moringa oleifera*

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The parts of *Moringa oleifera* were assessed for mineral composition and some heavy metal contents in this study, which included Ca, Mg, K, Na, Mn, Fe, Zn, Co, Se, Pb and Cd. Parts of the plant were obtained from Badagry in Lagos State, Nigeria. The samples were digested with HNO_3 and analysed for the mineral element content using atomic absorption spectrophotometer. The observed mean concentrations of the mineral elements were 26000, 643, 8210, 2980, 69.9, 169 and 15.3 mg/kg for Ca, Mg, K, Na, Mn, Fe and Zn, respectively. Co and Se, and the heavy metals Pb and Cd were not detected. There seemed to be no significant differences in the overall levels of the mineral elements in the different parts ($P > 0.05$). However, the observations showed that there were strong correlations between the levels of the elements in stem bark and root bark, stem bark and leaf, stem wood and pod, stem wood and seed, root bark and leaf, and pod and seed ($P < 0.01$); and between stem bark and root wood, root wood and root bark, and root wood and leaf ($P < 0.05$).

Key words: *Moringa oleifera*, mineral composition, heavy metals.

INTRODUCTION

Moringa oleifera commonly referred to as "Drumstick tree" belongs to the plant family *Moringaceae*. It is widely cultivated in semiarid, tropical and subtropical areas. The plant may be consumed as vegetable to improve nutriation, taken as medicinal plant to cure ailments and improve health, cultivated as forage for livestock, and used as live fencing.

Uses of various parts of *M. oleifera* plant have been reported. Powder produced from the seed is used as an effective primary coagulant for water treatment (Bhuptawat et al., 2007; Pritchard et al., 2010; Sánchez-Martín, 2010), and also possess the potential to remove cadmium from aqueous system (Sharma et al., 2006). The seed extract has been shown to have ameliorative effect on liver fibrosis in rats (Hamza, 2010), and also contain certain antitumor promoter (Guevara et al., 1999). Crude extracts and essential oil from *M. oleifera* possess anti-fungal activity against certain dermatophytes (Chuang et al., 2007). The leaves possess antioxidant properties (Verma et al., 2009). Some phytochemicals derived from the seeds possess insecticidal properties against mosquito (Prabhu et al., 2011).

This plant contains a considerable amount of various nutrients, and has been suggested as a good supplement for such nutrients as protein, fibre and minerals (Oduro et al., 2008; Jongrungruangchok et al., 2010). Also, including it in diets to supplement daily nutrient needs could help to fight against many diseases as nutraceuticals (Sharma et al., 2012). It is a viable supplement for dietary minerals (Aslam et al., 2005).

Several works have been done, reporting the levels of nutrients, including minerals in *M. oleifera,* with emphasis on variations in the levels as a function of geographical locations where the plant is grown; and the results

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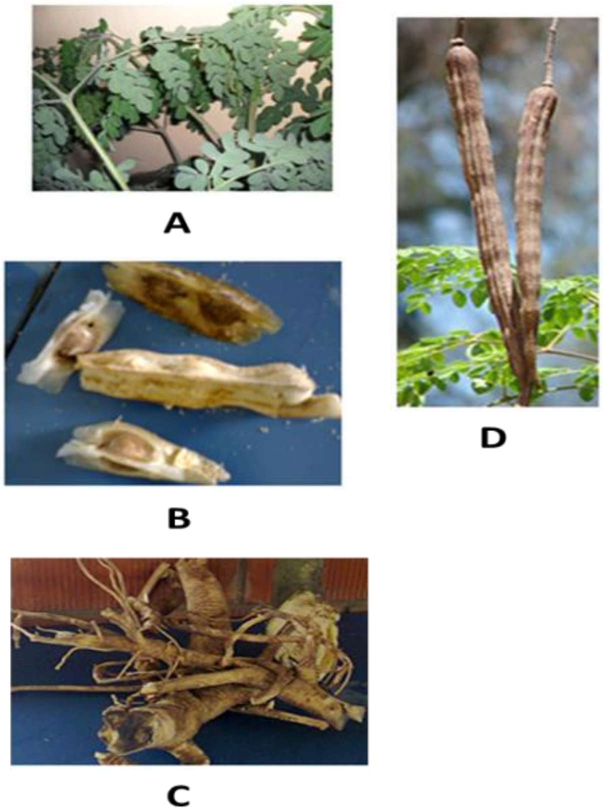

Figure 1. Parts of *M. oleifera* (A = leaf, B = seed, C = root, D = pod).

showed that there could be variations in the levels of macro and micro mineral elements in the plant, depending on the soil properties of the geographical locations where it grows (Anjorin et al., 2010). In the geographical location where this study is conducted, the most consumed part is the leaf, alleging that the needed nutrients are concentrated in the leaves. Reports on comparison of the levels of one element and the other in the different parts of the plant are scanty. Hence this study has these as objectives. The knowledge of the levels of various elements in the different parts of the plant may bring about efficient uses of other parts of the plant in addition to the leaves for the formulation as mineral supplement in both human and animal diet. The seeds are used in water purification; the knowledge of mineral composition of the seed will provide information on the elements that are likely to leach out into the water being purified or that may influence the water-purifying property. In addition, the study seeks to find out if relationships exist between the level of one element and the other, to provide an insight into some probable chemical interactions between the elements in the different parts of the plant. With this, one may be able to predict the level and/or behavior of one element by having the knowledge of the other. Others alleged that the parts like the root contain some toxins; but this is not within the objective of this study.

MATERIALS AND METHODS

The leaves (Figure 1A), root (Figure 1C) and stem of *M. oleifera* were obtained from a backyard at Badagry, Lagos state, while the pods (Figure 1D) together with the seeds (Figure 1B) and packaged powder for consumption (mainly produced from leaves) were obtained from University of Agriculture, Abeokuta. The leaves, roots and stem were thoroughly washed with water to minimize contaminations from adhering soil particles. The barks of the stem and the root were carefully peeled differently, while the woody parts were cut into tiny pieces to enhance quick drying. The samples were then air-dried for about four weeks in the laboratory. The dried samples were blended to fine particles, thoroughly cleaning the blender after each blend before the next. The woody parts, as these could not be blended, were cut into tiny particles with stainless scissors. The crucibles and glassware used were washed with detergents, acid solution and then rinsed with distilled water to avoid contaminations and thereby ensured the quality of the results. The samples were labeled SS (stem wood), SB (stem bark), RS (root wood), RB (root back), MP (powder), Lv (leaves), Pd (pod) and Sd (seed).

The analytical method used followed that described by Miroslav and Vladimir (1998) with slight modification. One gram of each sample dried to constant weight at 105°C was weighed in different porcelain crucibles, and then placed in a muffle furnace with the temperature slowly raised over 2 h to reach 550°C. They were left in the furnace at this temperature for 6 h. After cooling, the crucibles with the ash contents were removed from the furnace and allowed for further cooling. Five milliliters of 6 M nitric acid were added to each and carefully swirled to ensure complete dissolution. It was quantitatively filtered into a 100 ml volumetric flask and then made up to mark with distilled water. Blank was prepared in a similar way but omitting the plant sample.

The mineral elements, calcium (Ca), magnesium (Mg), potassium (K), sodium (Na), iron (Fe), zinc (Zn), manganese (Mn), Co and Se, as well as two toxic metals Pb and Cd, were determined using the atomic absorption spectrophotometer (AAS-Buck 205), at the wavelengths of 422.7, 285.2, 766.5, 589, 248.3, 213.9, 279.5, 240.7, 196, 283.3 and 228.9 nm, respectively, at the International Institute of Tropical Agriculture (IITA), Ibadan.

Using SPSS version 17, T-test was conducted on the results to see if there were differences in one set of the result and the other. The data were also analyzed using Pearson correlation analysis to determine if relationships exist. The analysis was done at 95% confidence level.

RESULTS

The observed mean concentrations of the mineral elements in *M. oleifera* plant (Table 1) were 26000, 643, 8210, 2980, 69.9, 169 and 15.3 mg/kg for Ca, Mg, K, Na, Mn, Fe and Zn, respectively. Co and Se, and the heavy metals Pb and Cd were not detected. Considering Figure 2, Ca (94900 mg/kg), Mg (762 mg/kg) and Na (9050 mg/kg) levels were highest in root bark, K (11300 mg/kg) in the seed, Mn (86 mg/kg) and Fe (214 mg/kg) in the leaves, and Zn (18 mg/kg) in powder and stem wood.

Using t-test, no significant difference was observed in the levels of Ca and K ($P = 0.321$), Ca and Na ($P = 0.095$) and Mg and Na ($P = 0.084$) at 95% confidence

Table 1. The distribution or spread of concentrations of the mineral elements and heavy metals in *M. oleifera*.

	Minimum statistic	Maximum statistic	Mean statistic standard error		Standard deviation statistic
Ca	1100	94900	26000	11000	31100
Mg	541	762	643	26	72
K	4750	11300	8210	747	2110
Na	696	9050	2980	1180	3320
Mn	63	86	70	3	7
Fe	154	214	169	7	20
Zn	11	18	15	1	2
*Co	-	-	-	-	-
*Se	-	-	-	-	-
*Pb	-	-	-	-	-
*Cd	-	-	-	-	-

*Not detected.

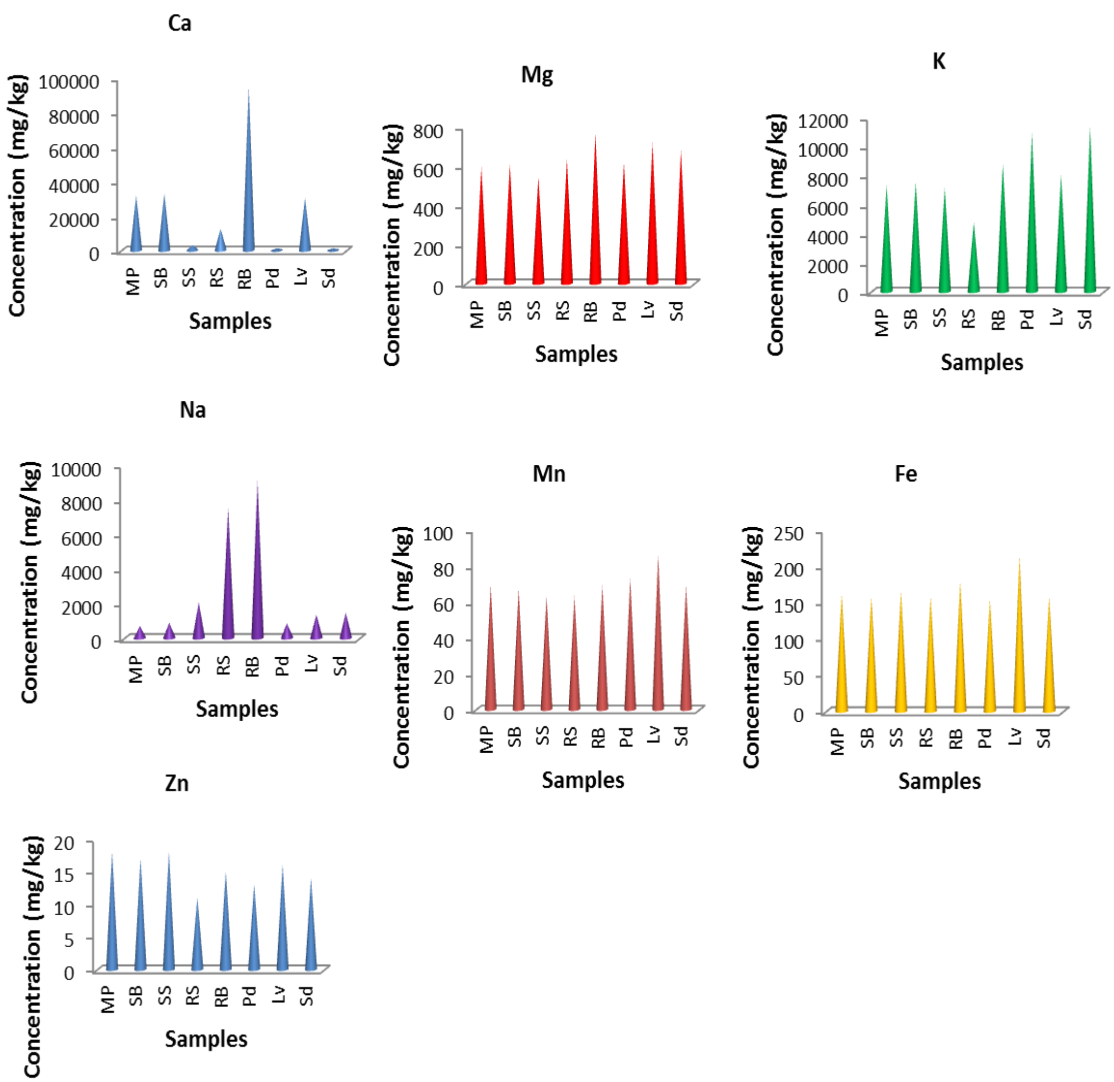

Figure 2. Concentration of mineral elements in MP (powder), SB (stem bark), SS (stem wood), RS (root wood), RB (root back), Pd (pod), Lv (leaves) and Sd (seed) of *M. oleifera*.

Table 2. Differences in the levels of the various mineral elements (paired samples test).

	Paired differences					t	df	Sig. (2-tailed)
	Mean	Std. deviation	Std. error mean	95% confidence interval of the difference				
				Lower	Upper			
Ca - Mg	13600	14900	5280	1110	26100	2.575	7	0.037
Ca - K	6040	16000	5650	-7320	19400	1.069	7	0.321
Ca - Na	11300	16500	5840	-2540	25100	1.929	7	0.095
Ca - Mn	14200	14900	5280	1690	26700	2.685	7	0.031
Ca - Fe	14100	14900	5280	1600	26600	2.667	7	0.032
Ca - Zn	14200	14900	5280	1750	26700	2.695	7	0.031
Mg - K	-7570	2090	740	-9320	-5820	-10.221	7	0.000
Mg - Na	-2340	3290	1160	-5090	411	-2.011	7	0.084
Mg - Mn	573	69	24	515	630	23.607	7	0.000
Mg - Fe	474	63	22	421	527	21.339	7	0.000
Mg - Zn	627	73	26	566	688	24.335	7	0.000
K - Na	5230	4530	1600	1440	9020	3.262	7	0.014
K - Mn	8140	2110	746	6370	9900	10.903	7	0.000
K - Fe	8040	2120	748	6270	9810	10.751	7	0.000
K - Zn	8190	2110	747	6430	9960	10.963	7	0.000
Na - Mn	2910	3330	1180	130	5690	2.475	7	0.043
Na - Fe	2810	3320	1170	34	5590	2.394	7	0.048
Na - Zn	2970	3330	1180	186	5750	2.523	7	0.040
Mn - Fe	-99	15	5	-111	-86	-18.970	7	0.000
Mn - Zn	55	8	3	48	61	20.442	7	0.000
Fe - Zn	153	20	7	137	170	22.271	7	0.000

level (Table 2), while others showed significant differences (at $P = 0.05$). Levels of Mn and Fe showed strong correlation (correlation coefficient of $r = 0.798$ and $P < 0.05$), using Pearson correlation analysis; there was little or no correlation between the levels of the others (Table 3): correlation coefficient between $r = -0.056$ and 0.574 ($0.136 \leq P \leq 0.895$). There seemed to be no significant differences in the overall levels of the mineral elements in the different parts of *M. oleifera* plants ($P > 0.05$) (Table 4). However, the observations showed that there were strong correlations (Table 5) between the levels of the elements in SB and RB, SB and Lv, SS and Pd, SS and Sd, RB and Lv, and Pd and Sd ($P < 0.01$); and between SB and RS, RS and RB and RS and Lv ($P < 0.05$).

DISCUSSION

The level of Ca was the highest (1100 to 94900 mg/kg). The highest level (94900 mg/kg) was observed in RB and the lowest (1100 mg/kg) in the pod. There was no significant difference between the levels of Ca and Na, and Ca and K ($P > 0.05$). There was little or no correlation between the levels of Ca and that of any of the other elements but Ca is a macro nutrient for humans, considerably sought for from other sources such as bones and shells of some crustaceans; if the level is this high, why is the part not commonly consumed? While the pod together with the seed is edible, there is restriction to the extent of human consumption of the root part of *M. oleifera* perhaps because of the information that it has some organic constituents likely to constitute neurotoxic effects (Gupta et al., 1999); however, further study may be needed to unfold the possibility of exploring the high Ca content for possible uses; or some possible food processing methods that may lead to detoxification and thereby making the root part consumable. The most consumed part is the leaf. The level of Ca observed in the leaves was 30300 mg/kg. The levels in literature (Jongrungruangchok et al., 2010) reporting similar works ranged from 15100 to 29510 mg/kg, which are comparable to those recorded in this study. This level of Ca in the leaf of *Moringa* is about four times that in milk (Wikipedia) and six times that observed in *Amaranthus* sp.(a common vegetable in Nigeria) (Sharma et al., 2012), hence as supplement in human diet, has the potential to meet the daily requirement. Ca is an important element in formation of bones and teeth; it is said to prevent osteoporosis (Susan and Lanham-New, 2008). Beneficial effects of Ca exist in the human body up to intake threshold of about 800 mg per day (Boon et al., 2005).

Table 3. Correlations between the concentration of one mineral element and the other in *M. oleifera*.

		Ca	Mg	K	Na	Mn	Fe	Zn
Ca	Pearson Correlation	1	-0.123	-0.439	-0.389	0.340	0.314	0.455
	Sig. (2-tailed)		0.771	0.276	0.340	0.410	0.448	0.257
Mg	Pearson Correlation	-0.123	1	0.293	0.504	0.515	0.574	-0.311
	Sig. (2-tailed)	0.771		0.482	0.203	0.191	0.136	0.453
K	Pearson Correlation	-0.439	0.293	1	-0.358	0.331	-0.073	-0.056
	Sig. (2-tailed)	0.276	0.482		0.384	0.424	0.864	0.895
Na	Pearson Correlation	-0.389	0.504	-0.358	1	-0.315	0.061	-0.479
	Sig. (2-tailed)	0.340	0.203	0.384		0.448	0.886	0.229
Mn	Pearson Correlation	0.340	0.515	0.331	-0.315	1	0.798*	0.072
	Sig. (2-tailed)	0.410	0.191	0.424	0.448		0.017	0.865
Fe	Pearson Correlation	0.314	0.574	-0.073	0.061	0.798*	1	0.227
	Sig. (2-tailed)	0.448	0.136	0.864	0.886	0.017		0.589
Zn	Pearson Correlation	0.455	-0.311	-0.056	-0.479	0.072	0.227	1
	Sig. (2-tailed)	0.257	0.453	0.895	0.229	0.865	0.589	

*Correlation is significant at the 0.05 level (2-tailed).

Table 4. Comparison of the concentration of mineral elements in the different parts of *M. oleifera* (paired samples test).

	Paired difference					t	df	Sig. (2-tailed)
	Mean	Standard deviation	Standard error mean	95% confidence interval of the difference				
				Lower	Upper			
MP - SB	-221	405	153	-596	154	-1.442	6	.199
MP - SS	3940	10900	4130	-6180	14100	.952	6	.378
MP - RS	2130	8060	3040	-5320	9570	.699	6	.511
MP - RB	-10400	23400	8840	-32100	11200	-1.181	6	.282
MP - Pd	3840	11900	4490	-7150	14800	.855	6	.425
MP - Lv	-13	726	274	-684	658	-.049	6	.963
MP - Sd	3680	11900	4520	-7370	14700	.814	6	.447
SB - SS	4160	11300	4280	-6320	14600	.971	6	.369
SB - RS	2350	8420	3180	-5440	10100	.738	6	.488
SB - RB	-10200	23000	8690	-31500	11000	-1.176	6	.284
SB - Pd	4060	12300	4640	-7280	15400	.876	6	.415
SB - Lv	208	1070	405	-782	1200	.513	6	.626
SB - Sd	3900	12300	4660	-7500	15300	.837	6	.435
SS - RS	-1810	4150	1570	-5650	2030	-1.154	6	.292
SS - RB	-14400	34200	12900	-46000	17300	-1.112	6	.309
SS - Pd	-96.4	1780	671	-174	1550	-.144	6	.890
SS - Lv	-3950	10300	3880	-13500	5550	-1.017	6	.348
SS - Sd	-260	1850	699	-1970	1450	-.373	6	.722
RS - RB	-12600	30800	11600	-41000	15900	-1.081	6	.321
RS - Pd	1710	5660	2140	-3520	6950	.802	6	.453
RS - Lv	-2140	7420	2800	-9000	4720	-.764	6	.474
RS - Sd	1550	5660	2140	-3680	6780	.725	6	.496
RB - Pd	14300	35200	13300	-18200	46800	1.075	6	.324
RB - Lv	10400	24000	9080	-11800	32600	1.149	6	.294
RB - Sd	14100	35200	13300	-18400	46700	1.061	6	.329
Pd - Lv	-3850	11200	4240	-14200	6510	-.910	6	.398

Table 4. Contd

Pd - Sd	-164	245	92	-390	62.1	-1.774	6	.126
Lv - Sd	3690	11300	4260	-6730	14100	.867	6	.419

SS (stem wood), SB (stem bark), RS (root wood), RB (root back), MP (powder), Lv (leaves), Pd (pod) and Sd (seed).

Table 5. Correlations between the concentrations of the mineral elements in one part of *M. oleifera* and the other.

		MP	SB	SS	RS	RB	Pd	Lv	Sd
MP	Pearson correlation	1	1.000**	.417	.846*	.986**	.137	.999**	.128
	Sig. (2-tailed)		.000	.352	.016	.000	.770	.000	.784
SB	Pearson correlation	1.000**	1	.417	.848*	.986**	.136	.999**	.127
	Sig. (2-tailed)	.000		.352	.016	.000	.771	.000	.785
SS	Pearson correlation	.417	.417	1	.532	.300	.935**	.449	.941**
	Sig. (2-tailed)	.352	.352		.219	.513	.002	.312	.002
RS	Pearson correlation	.846*	.848*	.532	1	.867*	.196	.858*	.216
	Sig. (2-tailed)	.016	.016	.219		.012	.673	.014	.642
RB	Pearson correlation	.986**	.986**	.300	.867*	1	-.009	.982**	-.013
	Sig. (2-tailed)	.000	.000	.513	.012		.985	.000	.978
Pd	Pearson correlation	.137	.136	.935**	.196	-.009	1	.169	.999**
	Sig. (2-tailed)	.770	.771	.002	.673	.985		.718	.000
Lv	Pearson correlation	.999**	.999**	.449	.858*	.982**	.169	1	.161
	Sig. (2-tailed)	.000	.000	.312	.014	.000	.718		.730
Sd	Pearson correlation	.128	.127	.941**	.216	-.013	.999**	.161	1
	Sig. (2-tailed)	.784	.785	.002	.642	.978	.000	.730	

**Correlation is significant at the 0.01 level (2-tailed); *Correlation is significant at the 0.05 level (2-tailed); SS (stem wood), SB (stem bark), RS (root wood), RB (root back), MP (powder), Lv (leaves), Pd (pod) and Sd (seed).

The concentrations of Mg ranged from 541 to 762 mg/kg, with the highest level in the RB as observed for Ca, and least in the stem wood. Mg and Ca have similar properties (same group in the periodic table of elements), and perhaps these influence the phyto-chemistry in the root of *M. oleifera*, and hence both are found at the highest level in the root. Further study is suggested to establish this. However, from the statistical analysis, there was significant difference in their levels (P < 0.05) and there was little or no correlation between their levels (r < 2.0 and P > 0.05). The level of Mg is not significantly different from that of Na (P > 0.05). The level in the most consumed part, the leaf, was 717 mg/kg, next to the level in the RB. These levels in the leaf are comparable to those reported (693 to 725 mg/kg) by Anjorin et al. (2010) for some Nigeria samples analyzed. Mg plays some roles in human health, namely, production and energy transport, contraction and relaxation of muscles, synthesis of protein and function of enzymes.

K levels ranged from 4750 to 11300 mg/kg, with the highest value in the seed and lowest in the root. If there are no anti-nutritional factors or other chemical components that may constitute toxins in the seed of this plant, the seed part may then be recommended as good source of potassium for supplements in diet. Concentration in the leaf was 8010 mg/kg, about three times higher than the level (3070 mg/kg) in *Ocimum gratissimum*, an indigenous green leafy vegetable in Nigeria (Thomas and Oyediran, 2008), and about three times higher than the levels in banana (Wikipedia). Yaméogo et al. (2011) reported K levels of 3086 to 22500 mg/kg in their study of chemical composition of *Moringa* leaf, and the value found in this study falls with this range. K helps in the proper function of the brain as well as nerves, thereby preventing stroke. It plays part in acid-base and water regulation in the blood and tissues. In contrast to Na, it has been reported that a high-potassium diet lowered blood pressure in individuals with raised blood pressure (He and MacGregor, 2008). In addition to its contribution to the electrolytes and function of the nerves in the human body, K has been linked to bone health and osteoporosis prevention according to studies of intake of 2500 mg/day done in United Kingdom, United States and Australia (Susan and Lanham-New, 2008). It may then be inferred from the results of this study that consumption of the leaf of *M. oleifera* could make up for the daily requirement of K in human diet. Observed further by Susan and Lanham-New (2008), the dietary content of pre-agricultural man suggests intake of K up to 7000 mg/day; hence, having the leaves of *M. oleifera* in the diet could bring the modern man to comparable level of diet as the pre-agricultural man.

The highest concentration of Na was observed in root bark. In all the parts studied, the levels ranged from 696 to 9050 mg/kg; and in the leaf, 1350 mg/kg. Observations of Na concentration were about the same for leaves and seeds. Aslam et al. (2005) reported similar levels (1292 to 1837 mg/kg) in M*oringa* leaves and 1032 to 2105 mg/kg in the pods. From this study and the references cited, it may be inferred that the level observed in the leaf is about twenty-three times higher than that in *Colocassia esculenta* (60 mg/kg) and about twelve times higher than the levels in *O. gratissimum* (113 mg/kg), two non-conventional indigenous green leafy vegetables in Nigeria (Thomas and Oyediran, 2008).

The concentrations observed for Mn in this study ranged from 63 to 86 mg/kg. This range is comparable to those reported (47.1 to 113.9 mg/kg) by Foidl et al. (2001). While there were significant differences between the levels of Mn and that of any of the other elements, strong correlation ($r = 0.798$, $P < 0.05$) existed between the levels of Mn and those of Fe. Mn helps in breaking down of fats, carbohydrates and proteins.

Fe level was highest in the leaves (214 mg/kg) and lowest in the pods (154 mg/kg). The leaves and fruit of *Moringa* are the most consumed, hence potential sources of Fe in human diet. Fe functions in the formation of haemoglobin and myglobin which are carriers of oxygen in the blood and blood vessels. Deficiency of Fe may result in anaemia. In determining the nutritional potential of *M. oleifera*, Oduro et al. (2008) obtained Fe level of 282.9 mg/kg in the leaf. For similar analysis, Jongrungruangchok et al. (2010) reported a range of 203.1 to 376 mg/kg. The level observed in this study is very much comparable to the levels in the latter reference.

Concentrations of Zn observed ranged from 11 to 18 mg/kg. It was 16 mg/kg in the leaf and 14 mg/kg in the seed. The levels are comparable to those recorded by Anjorin et al. (2010): 18 mg/kg in the leaf and 9.6 mg/kg in the seed. Zn enhances the function of immune system and proper functioning of some enzymes such as those involved in cell division, and growth. It has also been said to be involved in the phenomena of taste and smell.

Co, Se, Cd and Pb were not detected because the technique does not have a sufficient detection limit to quantify these elements in these types of samples.

Therefore, the graphite furnace and the generation hydride for selenium may be appropriate in future determinations.

Conclusion

No significant difference was observed in the levels of Ca and K, Ca and Na, and Mg and Na at 95% confidence level, while others showed significant differences. Levels of Mn and Fe showed strong correlation but there was little or no correlation between the levels of the others. There seemed to be no significant differences in the overall levels of the mineral elements in the different parts of *M. oleifera* plants. However, the observations showed that there were strong correlations between the levels of the elements in stem bark and root bark, stem bark and leaf, stem wood and pod, stem wood and seed, root bark and leaf, and pod and seed; and between stem bark and root wood, root wood and root bark, and root wood and leaf.

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