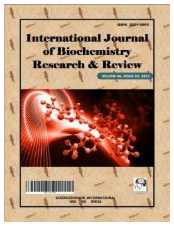

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Assessment of Levels of Nutrients in Selected ALVs at Different Harvesting Stages and Locations of Production in Western Kenya

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Proper nutrition contributes to declines under-five mortality rates and improves the productivity of adults. Addressing nutritional problems requires adequate information on the diets of individuals and populations. African leafy vegetables (ALVs) are widely consumed and often harvested at different stages after planting with the help of different communities. Four ALVs namely *Vigna unguiculata*, *Amaranthus hybridus*, *Cleome gynandra* and *Solanum scabrum* are commonly grown vegetables in western Kenya, their potentials have not been evaluated to supply the nutrients. However, nutritional values may vary depending on the species, harvesting stage and location of production. The effects of species, harvesting stages and location of production on the nutritional value of selected ALVs were evaluated. The trials were laid out in a randomized complete block design in three replicates in Busia, Kisumu, and Lela. Leaves were sampled at different harvesting stages and analyzed for N, P, K, Na, Ca, Mg, Mn, Fe, and Zn levels. *Amaranthus hybridus* had significantly ($P \leq 0.05$) higher levels of P, Ca, Zn, Mn, and Na. The N, P, K, Ca, Mg, Zn and Fe levels significantly ($P \leq 0.05$) increased then decreased with harvesting stage. The levels of nutrients significantly ($P \leq 0.05$) varied with location of production except for Na. The ALVs from Kisumu site had significantly ($P \leq 0.05$) higher

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levels of N, P, K, Ca, Mg and Zn, ALVs from Lela site had higher levels of Mn and Na while ALVs from Busia site had higher levels of Fe. *Amaranthus hybridus* is a better contributor of-of P, Ca, Zn, Mn, and Na. The Fe, Mg and Zn levels were above the Recommended Dietary Allowance and may be used to alleviate deficiencies associated with such nutrients. Harvesting the ALVs at the stage the nutrients attain their optimum levels is recommended. There is a need for the development of regional food composition tables for all ALVs in Kenya.

Keywords: African leafy vegetables; harvesting stages; locations of production; nutrients; recommended dietary allowances; western; Kenya.

1. INTRODUCTION

Over the past several decades, sub- Saharan Africa has been experiencing a nutrition transition in which traditional foods and food habits have been progressively replaced by the globalized food culture of the multinational corporations [1] The nutrition transition has significantly resulted in changes in diet and lifestyle habits [2]. Such changes have led to a paradoxical phenomenon seen in developing countries where under nutrition and over nutrition coexist [2]. Kenya, like other developing countries in sub-Saharan Africa, is faced with this double-burden [3]. Under nutrition comprise of a number of nutritionally related conditions such as protein-energy malnutrition and micronutrient deficiencies. Increasing trends in child overweight have been noted in most world regions, not only developed countries, where prevalence is highest (15% in 2011) and 7% in Africa [4].

Estimates of the prevalence of malnutrition in children less than five years of age and women of reproductive age showed a large variation by province [5], reflecting the considerable variability in environmental and socioeconomic risk factors [6]. Many Kenyans living in western Kenya suffer from nutrient deficiency diseases [7] with an under-five mortality rate of 206 per 1000 live births in Nyanza province [5]. Poverty normally manifests itself mainly through malnutrition and poor health. The poverty incidence showed a large variation by province, with Western and Nyanza all above 45% of the population [8].

Proper nutrition contributes significantly to declines in under-five mortality rates [4] and also improves the productivity of adults. The problems of micronutrient malnutrition and over nutrition cannot be addressed without adequate information on the diets of individuals and populations. This in turn requires reliable data on both the consumption of foods and their nutrients contents.

Kenya is endowed with many varieties of indigenous food plants [9] like ALVs that can have outstanding potential to supply RDAs of relevant nutrients. Many Kenyans especially people from western regions consume ALVs together with other starchy food stuff and other proteins for general meals [10]. It is their general believes that the vegetables are important sources of vitamins and minerals (Fe and Ca) [11,12,13]. However, the levels of beneficial nutrients in the ALVs grown in western Kenya have not been documented to guide consumption levels to supply RDAs for relevant nutrients. Generally, poor communities that cannot afford expensive nutrient-dense animal source foods consume a lot of ALVs which are freely available or are cheap [14,15]. Many affluent adults are also now resolving to indigenous foods [16], as they are believed to be nutritious.

Food composition data is essential in nutritional research for assessing and planning interventions involving national food and nutrition policies and prescribing therapeutic and institutional diets for individuals and populations. It is essential that efforts to analyze the nutrient content of foods are continued in African countries. Up-dated information is needed for all types of foods including ALVs. Foods, being biological materials, exhibit variations in nutrient levels due to factors such as cultivar and location of production [17] Harvesting stage and environmental conditions [18,19,20] have also been established to influence nutritional value in ALVs. It is therefore important to determine the optimal harvesting stage for which the selected ALVs would exhibit optimal nutrients for each location of production. The data may be used to develop regional food composition table for the selected ALVs in western Kenya. The ALVs composition table developed by Sehmi [21] for Kenyan ALVs is questionable as samples were bought from different markets.

Minerals uptake in plants is through the root system from the soil solution [22,23]. Soil compositions tend to vary widely as evidenced by their texture and color [24]. Fluctuations in environmental factors such as temperature and soil moisture can affect the mineral nutrient content of plant leaves considerably [25]. These factors influence both the availability and uptake of nutrients by the roots and the shoot growth rate.

Nutrient content in vegetables varies according to their availability in the soil at different collection sites and plant uptake [26]. The nutritional values of vegetables differ not only according to type of vegetable but also according to the geographical area of production [27]. Zinc and Fe levels in *Cleome gynandra* and *Amaranthus hybridus* varied in Kongwa, Muheza and Arumeru districts in Tanzania [27]. Iron and Zn levels in *Corchorus olitorius* and *Corchorus trilocularis* in Muheza district were lower than those in Morogoro district in Tanzania [28]. Heavy metal content significantly varied in ALVs planted in different locations in urban and peri-urban Nairobi, Kenya [29]. No study has been done to establish variations in nutrient levels in ALVs in different locations of western Kenya.

Western Kenya is known to produce *Vigna unguiculata*, *Amaranthus hybridus*, *Cleome gynandra* and *Solanum scabrum* almost throughout the year due to favorable soils and climatic conditions, which vary slightly across the region spanning from the Lake Victoria to high equatorial rainy regions [24]. These variations may lead to variations in nutrients uptake by plants. Studies have been conducted on several foods and cash crops except for the naturally occurring ALVs.

ALVs can be harvested at different stages of plant growth, ranging from young seedlings to the late juvenile stage [30], but data on the changes in leaf nutritional value with plant age are scanty. The nutritive qualities of ALVs are significantly affected by plant age [31,32] The yields, beta-carotene and phenolic compounds increase then decrease [33] while crude proteins levels do not change significantly with age. Usually higher harvesting frequency results in a higher leaf yield than a single harvest [31,32] however, it's not known how this affects mineral distribution. Variations in the levels of nutrients in ALVs at different harvesting stages had been reported in Nairobi, Kenya [34], Zimbabwe [35] and South Africa [30]. Highest levels of Ca and Zn in *Amaranthus cruentus* were attained at 3 WAE, K at 6 WAE and P at 4 WAE in Zimbabwe [35] and South Africa [30]. However, highest levels of Na were reported at 6 WAE in Zimbabwe [35] and 7 WAE in South Africa [30]. It is not known how harvesting stage affects the distribution of nutrients in ALVs grown in western Kenya.

2. METHODOLOGY

2.1 Planting of ALVs

The trials were laid down in three geographical locations of Busia (altitude: 1350 m above mean sea level, 0º28´N 34º6´E), Kisumu (altitude: 1132 m above mean sea level, 0º03´S 34º45´E) and Lela (altitude: 1560 m above sea level, 0º1´S 34º36´E) in western Kenya from April to September, 2009. The experiment was laid down in a randomized complete block design (RCBD) replicated three times. Each replicate (4 m2) was divided into four plots (each 1 m2) to cater for the four ALVs species namely: *Cleome gynandra,* variety PS*, Solanum scabrum,* variety SS49*, Amaranthus hybridus,* amine variety and *Vigna unguiculata,* fahari variety which was distributed randomly. Land preparation was done by clearing the weeds followed by deep ploughing. Harrowing was done using a folk and leveled with a rake. Certified seeds (from Lagrotech, Kisumu) were mixed with the soil in the ratio 1:10 and saw directly 2 cm deep in rows with an inter-row spacing of 30 cm (planting density 11 plants m-2, i.e. 30 cm by 30 cm) [36], and covered with a thin layer of soil. Weeding was carried out three times; the 1st on the 3rd, 2nd on the 5th and 3rd on the 7th week, while thinning was done with an inter-plant spacing of 30 cm on the 3rd week just before weeding and spraying done using Ortus 5 SC (fenpyroximate as the active ingredient) for insect and pathogen control.

2.2 Preparation of Leaf Samples

The upper palatable aerial parts of individual plants (2nd and 3rd leaves) were sampled fortnightly four times, after 4 WAE. The Association of Official Analytical Chemists [37] method of sample preparation was used. Leaf samples were washed in deionized water three times to remove soil particles, oven-dried at 60ºC and crushed into a fine powder using a mill. Dried and ground leaves (0.5 g) in porcelain crucible were ashed in a muffle furnace at 450ºC until greyish white ash was obtained. The samples were cooled on top of the asbestos sheet and 5 mL analytical grade 1 N HNO3 solution added to each sample for digestion. This was evaporated to dryness on a steam bath to ensure complete digestion. The samples were returned to the furnace for a further 15 minutes until a perfect grey ash was obtained. The grey ash was cooled on asbestos sheet and 10 mL of analytical grade 1 N HCl added for mineral extraction then filtered into 50 mL volumetric flask. The crucible and filter paper were rinsed three times and the aliquots added to the flask before making to the mark with 0.1 N HCl. The extract was analyzed for N, P K, Na, Ca, Mg, Mn, Fe, and Zn.

2.3 Sampling and Preparation of Soil Samples

Soil samples at depths of 0-15 and 15- 30 cm were sampled using Johnson Bucket soil auger (Thompson type) in the experimental plots in a diagonal pattern [38] before the seeds were planted to determine the soil nutrient status at the time the experiment commenced for soil pH and N, P, K, Na, Ca, Mn, Mg, Fe, and Zn analyses. Total K, Na, Ca, Mg, Mn, Fe, P, and Zn were determined by digestion of 5.0 g (composite samples) of the dried soil (passed through 2 mm mesh size) using 25 mL of a 3:1 mixture of concentrated HNO3 and HCl (analytical reagent grade, from Kobian, Kenya), in a 50 mL polythene bottle. Activated charcoal (0.5 g, analytical grade, from Kobian, Kenya) was added to remove color. The mixture was a stopper, mixed well and shaken mechanically for six hours followed by filtration through medium speed filter paper into a 50 mL volumetric flask and made to mark with 0.1 N HCl. The elements N, P K, Na, Ca, Mg, Zn, Fe, and Mn, were analyzed as explained in leaf samples.

2.4 Determination of N, P, K, Na, Ca, Mg, Zn, Fe, and Mn in Leaf and Soil Samples

The leaf and soil samples were pipetted (0.5 mL) into 50 mL volumetric flasks and made to the mark with double deionized water after addition of 1 mL strontium chloride (analytical grade, SrCl2.6 H2O from Kobian, Kenya). The purpose of strontium chloride was to remove interference in the absorption of the specific metal by other metals at the same wavelength by acting as a buffer hence preventing further ionization of the ions [39]. The extract was analyzed for K, Na and Ca using flame photometer (Jenway Model, PFP 7) and Mg, Mn, Fe, and Zn using atomic absorption spectrometer, AAS (Model AA- 6200, Shimadzu, Corp., Kyoto, Japan). Wavelengths of 285.2 nm, 279.5 nm 248.3 nm, and 213.9 nm was used to measure the absorbance of Mg, Mn, Fe, and Zn, respectively. Analytical grade salts of KCl, NaCl, CaCO3, KMnO4, and metals: Fe, Mg, and Zn (from Kobian, Kenya) were used to prepare known concentrations per salt in 100 mL volumetric flasks after addition of 1 mL strontium chloride. These salts were used as standards and a calibration curve was drawn which was used to determine the concentration of the analyze. Total phosphates and N were determined by a method described by [40] and AOAC [41] respectively with slight modifications. Absorbance of P was measured using an ultraviolet spectroscopy (PU-8625 UV-Vis spectrophotometer) of the yellow reaction product, vanadomolybdophosphate, at 460 nm.

2.5 Determination of Soil pH

Soil pH in water (2.5:1 water to soil ratio) was determined using glass-electrode pH meter (3071 Jenway) according to Rhoades (1982). The pH meter was calibrated with pH 7.0, pH 4.0 buffer before use. In this method, 50 mL of deionized water was added to 20 g of air-dried soil (passed through 2 mm mesh size) into a 50 mL beaker. The mixture was stirred for 10 minutes and allowed to stand for 30 minutes. The mixture was again stirred for 2 minutes and the pH reading of the partly settled suspension was taken. The pH electrode was rinsed with deionized water and wiped dry with filter paper after each reading.

2.6 Data Analysis

The data for ALVs was analyzed using RCBD in a 3-factorial arrangement with species as the main treatment, location as sub- treatment, and harvesting stage as sub-sub- treatment while soil data was analyzed using RCBD in a 1-factorial arrangement using MSTAT-C statistical package (Michigan State University, MI) for ANOVA.

3. RESULTS AND DISCUSSION

3.1 Effect of Location of Production on Levels of Nutrients in Selected ALVs

The levels of N, P, K, Ca, Mg, Fe, Zn and Mn significantly ($P \leq 0.05$) varied with location of production except for Na that showed non-significant variation (Tables 1-9).

Table 1. Variation in levels of N (%) in selected ALVs at different locations and harvesting stages

Loc	H.stage (wks)	Species				M. stage	M. loc
		V. unguiculata	*A. hybridus*	*C. gynandra*	*S. scabrum*		
	4	2.241	1.735	1.641	2.019	1.909	
Busia	6	2.570	1.743	1.644	2.125	2.020	
	8	2.546	1.749	1.659	2.227	2.045	
	10	2.290	1.757	1.664	2.365	2.019	
	Mean spp	2.412	1.746	1.652	2.184		1.998
	CV (%)			4.980			
	LSD (P≤0.05)			0.129		0.129	
	Interaction			0.184			
Kisumu	4	2.365	1.775	1.766	2.214	2.030	
	6	2.552	1.872	1.769	2.345	2.135	
	8	2.576	1.878	1.778	2.431	2.166	
	10	2.577	1.837	1.793	2.563	2.193	
	Mean species	2.518	1.841	1.777	2.388		2.131
	CV (%)			2.090			
	LSD (P≤0.05)			0.058		0.058	
	Interaction			0.082			
Lela	4	1.538	1.591	1.554	1.786	1.617	
	6	1.579	1.634	1.565	1.825	1.651	
	8	1.585	1.642	1.571	1.768	1.641	
	10	1.587	1.650	1.574	1.845	1.664	
	Mean species	1.572	1.629	1.566	1.806		1.643
	CV (%)			3.140			
	LSD (P≤0.05)			0.067		NS	
all sites	4	2.048	1.700	1.654	2.006	1.852	
	6	2.234	1.750	1.660	2.098	1.935	
	8	2.236	1.756	1.669	2.142	1.951	
	10	2.156	1.748	1.677	2.258	1.959	
	Mean species	2.167	1.739	1.665	2.126	1.924	1.924
	CV (%)			3.620			
	LSD (P≤0.05)			0.052		0.052	0.061
	Interaction			0.120			

Table 2. Variation in levels of P (mg/100 g DW) in selected ALVs at different locations and harvesting stages

Loc	H. stage (wks)	Vegetable species				M. stage	M. loc
		V. unguiculata	*A. hybridus*	*C. Gynandra*	*S. scabrum*		
	4	18.679	48.370	26.726	37.363	32.784	
Busia	6	23.097	52.128	29.583	42.010	36.704	
	8	19.642	48.340	25.293	36.798	32.518	
	10	16.988	42.790	21.215	35.432	29.107	
	Mean spp	19.602	47.907	25.704	37.901		32.778
	CV (%)			4.120			
	LSD (P≤0.05)			1.756		1.756	
	Interaction			NS			
Kisumu	4	35.057	71.794	41.292	60.946	52.356	
	6	39.505	77.158	46.168	64.626	56.864	
	8	33.255	73.958	41.292	58.370	51.719	
	10	32.170	69.404	38.414	52.693	48.170	
	Mean spp	34.997	73.078	41.875	59.159		52.277
	CV (%)			6.920			
	LSD (P≤0.05)			4.696		4.696	
	Interaction			6.677			
Lela	4	12.473	18.876	16.760	17.002	16.278	
	6	13.089	21.483	16.150	19.113	17.459	

Loc	H. stage (wks)	Vegetable species					
		V. unguiculata	*A. hybridus*	*C. Gynandra*	*S. scabrum*	M. stage	M. loc
	8	10.331	16.821	13.697	16.658	14.377	
	10	8.440	17.195	12.749	18.328	14.178	
	Mean spp	11.083	18.594	14.839	17.775		15.573
	CV (%)			4.610			
	LSD (P≤0.05)			0.933		0.933	
	Interaction			1.327			
all sites	4	22.070	46.343	28.371	38.437	33.806	
	6	25.230	50.256	30.634	41.916	37.009	
	8	21.076	46.373	26.761	37.275	32.871	
	10	19.199	43.130	24.126	35.485	30.485	
	Mean spp	21.894	46.526	27.473	38.278		33.543
	RDA (mg)						1200.000
	AVC*						3.577*
	CV (%)			7.140			
	LSD (P≤0.05)			1.795		1.795	2.102
	Interaction			4.106			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Table 3. Variation in levels of K (mg/100 g DW) in selected ALVs at different locations and harvesting stages

Loc	H. stage (wks)	Species					
		V. unguiculata	*A. hybridus*	*C. Gynandra*	*S. scabrum*	M. stage	M. loc
	4	69.932	80.402	69.337	65.480	71.288	
Busia	6	76.472	83.000	79.916	75.475	78.715	
	8	65.550	80.747	76.190	70.085	73.143	
	10	62.472	71.944	68.808	58.303	65.381	
	Mean spp	68.607	79.023	73.563	67.335		71.236
	CV (%)			9.480			
	LSD (P≤0.05)			8.771		8.771	
	Interaction			NS			
Kisumu	4	95.271	127.603	102.787	100.574	106.559	
	6	102.453	131.017	112.647	109.503	113.905	
	8	92.941	125.010	98.092	98.118	103.540	
	10	91.227	118.077	89.693	92.176	97.793	
	Mean spp	95.493	125.427	100.804	100.093		105.449
	CV (%)			1.360			
	LSD (P≤0.05)			1.868		1.868	
	Interaction			2.655			
Lela	4	43.679	56.681	42.735	49.659	48.189	
	6	47.607	59.085	47.297	53.162	51.788	
	8	41.521	56.348	43.078	49.633	47.645	
	10	35.480	52.679	39.967	43.049	42.794	
	Mean spp	42.072	56.198	43.269	48.876		47.604
	CV (%)			3.130			
	LSD (P≤0.05)			1.933		1.933	
	Interaction			2.749			
all sites	4	69.627	88.229	71.620	71.904	75.345	
	6	75.511	86.253	79.953	79.380	80.274	
	8	66.670	87.368	72.453	72.612	74.776	
	10	63.060	80.900	66.156	64.509	68.656	
	Mean spp	68.717	85.687	72.546	72.101		74.763
	RDA (mg)						2000.000
	AVC*						2.675*
	CV (%)			5.500			
	LSD (P≤0.05)			3.081		3.081	3.608
	Interaction			NS			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Table 4. Variation in levels of Ca (mg/100 g DW) in selected ALVs at different locations and harvesting stages

	Species					
H. stage (wks)	*V. unguiculata*	*A. hybridus*	*C. Gynandra*	*S. scabrum*	M. stage	M. loc
4	178.370	239.350	180.327	185.643	195.923	
6	190.890	254.447	254.293	192.570	223.050	
8	155.707	208.777	170.740	175.203	177.607	
Mean species	168.597	222.485	190.424	174.710	159.637	189.054
CV (%)			0.830			
LSD (P≤0.05)			2.032		2.032	
Interaction			2.886			
4	279.930	401.780	198.053	295.963	252.015	
6	287.653	435.767	227.993	315.327	408.048	
8	240.133	404.617	188.367	247.210	196.294	
10	200.343	390.027	170.763	211.883	267.596	
Mean species	252.015	408.048	196.294	267.596		280.988
CV (%)			1.360			
LSD (P≤0.05)			49.391		49.391	
Interaction			70.222			
4	106.937	141.590	148.107	120.100	129.183	
6	116.290	156.247	173.353	133.480	144.842	
8	83.264	125.120	138.480	110.390	114.321	
10	72.890	100.038	122.030	104.967	99.981	
Mean species	94.845	130.749	145.500	117.234		122.082
CV (%)			2.310			
LSD (P≤0.05)			3.668		3.668	
Interaction			5.215			
4	188.412	260.907	175.496	200.569	206.346	
6	198.278	282.153	218.547	213.792	228.193	
8	159.701	246.171	165.872	177.601	187.336	
10	140.884	225.811	149.710	154.091	167.624	
Mean species	171.819	253.760	177.406	186.513		197.375
RDA (mg)						1000.00
AVC*						0.506*
CV (%)			12.140			
LSD (P≤0.05)			17.968		17.968	21.043
Interaction			NS			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Table 5. Variation in levels of Mg (mg/100 g DW) in selected ALVs at different locations and harvesting stages

Loc	H. stage (wks)	Vegetable species					
		V. unguiculata	*A. Hybridus*	*C. gynandra*	*S. scabrum*	M. stage	M. loc
	4	28.823	17.807	34.527	366.743	111.975	
Busia	6	31.523	19.990	37.397	390.283	119.798	
	8	34.103	22.380	29.270	424.707	127.615	
	10	32.920	22.690	26.747	434.347	129.176	
	Mean spp	31.842	20.717	31.985	404.020		122.141
	CV (%)			1.440			
	LSD (P≤0.05)			2.287		2.287	
	Interaction			3.251			
Kisumu	4	44.123	25.037	54.190	569.177	173.132	
	6	44.780	25.993	56.103	577.193	176.018	
	8	48.607	24.317	49.323	583.237	176.371	
	10	46.160	23.403	45.700	573.010	172.068	
	Mean spp	45.917	24.687	51.329	575.654		174.397
	CV (%)			1.330			
	LSD (P≤0.05)			3.020		3.020	
	Interaction			4.294			

Table 5. Continued.

Lela	4	22.147	18.863	29.543	268.913	84.867	
	6	26.413	17.467	32.277	281.163	89.330	
	8	25.510	19.707	26.093	327.047	99.589	
	10	22.487	18.623	22.470	309.103	93.171	
	Mean spp	24.139	18.665	27.596	296.557		91.739
	CV (%)			4.290			
	LSD (P≤0.05)			5.114		5.114	
	Interaction			7.272			
all sites	4	31.698	20.569	39.420	401.611	123.324	
	6	34.239	21.150	41.926	416.213	128.382	
	8	36.073	22.134	34.896	444.997	134.525	
	10	33.856	21.572	31.639	438.820	131.472	
	Mean spp		33.966	21.356	36.970	425.410	129.426
	RDA (mg)						350.000
	AVC*						0.270*
	CV (%)			2.160			
	LSD (P≤0.05)			2.099		2.099	2.458
	Interaction			4.802			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Table 6. Variation in levels of Fe (mg/100 g DW) in selected ALVs at different locations and harvesting stages

Loc	H.stage (wks)	Vegetable species					
		V. unguiculata	***A. hybridus***	***C. gynandra***	***S. scabrum***	**M. stage**	**M. loc**
	4	9.413	9.079	15.304	9.100	10.724	
Busia	6	10.022	10.562	15.894	9.248	11.432	
	8	10.206	10.929	16.556	9.346	11.759	
	10	10.454	11.406	15.499	9.436	11.699	
	Mean species	10.024	10.494	15.813	9.282		11.403
	CV (%)			5.400			
	LSD (P≤0.05)			0.799		0.799	
	Interaction			NS			
Kisumu	4	7.564	6.835	13.929	5.594	8.481	
	6	8.052	7.674	15.764	6.298	9.447	
	8	8.087	7.848	15.000	6.514	9.362	
	10	8.102	10.518	16.040	6.691	10.338	
	Mean species	7.951	8.218	15.183	6.274		9.407
	CV (%)			8.470			
	LSD (P≤0.05)			1.035		NS	
	Interaction			1.472			
Lela	4	6.046	5.445	7.688	3.392	5.643	
	6	6.517	5.555	9.491	3.741	6.326	
	8	6.622	5.641	9.367	3.931	6.390	
	10	6.880	6.082	9.053	4.260	6.569	
	Mean species	6.516	5.681	8.900	3.831		6.232
	CV (%)			7.120			
	LSD (P≤0.05)			0.576		0.576	
	Interaction			NS			
all sites	4	7.675	7.119	12.307	6.029	8.282	
	6	8.197	7.930	13.716	6.429	9.068	
	8	8.305	8.139	13.641	6.597	9.170	
	10	8.479	9.335	13.531	6.796	9.535	
	Mean species	8.164	8.131	13.299	6.463		9.014
	RDA (mg)						18.000
	AVC*						0.199*
	CV (%)			7.090			
	LSD (P≤0.05)			0.480		0.480	0.562
	Interaction			NS			

*AVC *, the average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA.*

Table 7. Variation in levels of Zn (mg/100 g DW) in selected ALVs at different locations and harvesting stages

		Species					
Loc	**H.stage (wks)**	***V. unguiculata***	***A. Hybridus***	***C. gynandra***	***S. scabrum***	**M. stage**	**M. loc**
	4	1.554	2.065	1.531	1.795	1.736	
Busia	6	1.715	2.266	1.800	1.963	1.936	
	8	1.615	2.045	1.382	1.675	1.679	
	10	1.368	1.922	1.032	1.624	1.486	
	Mean species	1.563	2.075	1.436	1.764		1.710
	CV (%)			3.780			
	LSD (P≤0.05)			0.084		0.084	
	Interaction			0.119			
Kisumu	4	1.098	2.973	2.371	1.609	2.013	
	6	1.177	3.871	2.676	1.883	2.402	
	8	1.058	3.150	2.316	1.028	1.888	
	10	0.910	2.887	1.926	0.882	1.651	
	Mean species	1.061	3.220	2.322	1.350		1.988
	CV (%)			2.220			
	LSD (P≤0.05)			0.057		0.057	
	Interaction			0.081			
Lela	4	0.636	1.192	0.887	0.946	0.915	
	6	0.759	1.278	0.995	1.182	1.053	
	8	0.612	1.221	0.830	0.806	0.867	
	10	0.464	1.076	0.687	0.685	0.728	
	Mean species	0.618	1.192	0.850	0.905		0.891
	CV (%)			5.060			
	LSD (P≤0.05)			0.058		0.058	
	Interaction			0.083			
all sites	4	1.096	2.077	1.596	1.450	1.555	
	6	1.217	2.472	1.824	1.676	1.797	
	8	1.095	2.139	1.509	1.169	1.478	
	10	0.914	1.962	1.215	1.064	1.289	
	Mean species	1.080	2.162	1.536	1.340		1.530
	RDA (mg)						9.000
	AVC*						0.588*
	CV (%)			3.470			
	LSD (P≤0.05)			0.037		0.037	0.047
	Interaction			0.091			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

The ALVs from Kisumu site had significantly (P≤0.05) higher levels of N, P, K, Ca, Mg and Zn, ALVs from Lela site had higher levels of Mn and Na while ALVs from Busia site had higher levels of Fe.

Variation in levels of nutrients with the location of production had also been observed earlier in Tanzania. The Fe and Zn levels in amaranths, African nightshades, and cowpea significantly (P≤0.05) varied in Kongwa, Singida and Muheza [27] and in Iringa and Morogoro districts in Tanzania [28].

The variations in this study may be attributed to variations in initial soil pH and soil nutrient levels. Soil analysis was done to ascertain soil nutrient status especially N, P and K, primary nutrients required in relatively large amounts [42]. This was done to establish if the levels of nutrients in the soil could support plant growth as most small holder farmers grow ALV without fertilizer application and soil nutrient replenishment. On the other hand, excess nutrients in the soil-crop system increases the risk of nutrient losses to the environment and the risk of impaired product quality [43,44], whereas deficiencies reduce crop production.

Initial soil nutrient levels and pH significantly (P≤0.05) varied in the three geographical locations (Table 10) Soils from Busia and Kisumu sites were nearly at neutral pH while those from Lela were slightly acidic.

Table 8. Variation in levels of Mn (mg/100 g DW) in selected ALVs at different locations and harvesting stages

		Vegetable species					
Loc	**H.stage (wks)**	***V. unguiculata***	***A. Hybridus***	***C. gynandra***	***S. scabrum***	**M. stage**	**M. loc**
	4	0.018	0.055	0.026	0.028	0.032	
Busia	6	0.029	0.044	0.021	0.026	0.030	
	8	0.038	0.045	0.031	0.016	0.032	
	10	0.041	0.039	0.024	0.025	0.032	
	Mean species	0.031	0.046	0.025	0.024		0.032
	CV (%)			22.910			
	LSD (P≤0.05)			0.009		NS	
	Interaction			0.013			
Kisumu	4	0.021	0.065	0.046	0.045	0.044	
	6	0.036	0.084	0.045	0.036	0.050	
	8	0.047	0.084	0.034	0.033	0.050	
	10	0.051	0.089	0.019	0.032	0.048	
	Mean species	0.039	0.081	0.036	0.036		0.048
	CV (%)			28.18			
	LSD (P≤0.05)			0.018		0.018	
	Interaction			0.025			
Lela	4	0.035	0.085	0.052	0.088	0.065	
	6	0.042	0.089	0.051	0.094	0.069	
	8	0.056	0.085	0.057	0.099	0.074	
	10	0.065	0.085	0.062	0.100	0.078	
	Mean species	0.050	0.086	0.056	0.095		0.072
	CV (%)			7.580			
	LSD (P≤0.05)			0.007		0.007	
	Interaction			0.009			
all sites	4	0.025	0.068	0.041	0.054	0.047	
	6	0.036	0.072	0.039	0.052	0.050	
	8	0.047	0.071	0.041	0.049	0.052	
	10	0.052	0.071	0.035	0.053	0.053	
	Mean species	0.040	0.071	0.039	0.052		0.051
	RDA (mg)						1.800
	AVC*						3.529*
	CV (%)			18.430			
	LSD (P≤0.05)			0.007		0.007	0.008
	Interaction			0.016			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Low soil pH affects different physiological and biochemical processes, both in the soil and plant [45]. For instance, low soil pH enhances the uptake of Mn, [46,47] and Zn [48] while high soil pH enhances the uptake of K, Na, Ca, and Mg [49,50] by plants. The uptake of K, Ca and Mg by ALVs were enhanced with near neutral soil pH in Kisumu and Busia sites. This was in agreement with previous studies but contrary to P and Na intake. Similarly, the decrease in the uptake of Mn with enhanced pH in the current study was in agreement with previous studies [46,47].

The soil nutrient levels significantly (P≤0.05) varied in the three sites except for N that showed non-significant variation. These findings were in agreement with observations made earlier in different locations of western Kenya [24]. Soils from Kisumu site had significantly (P≤0.05) higher levels of N, P, K, Ca, Mg, and Zn; soils from Lela site had lower levels but with higher levels of Mn and Na while soils from Busia site had higher levels of Fe. The total N, K, Ca, and Mg levels in soils from the three sites were very high compared to [51] ratings. However, ALVs from Lela site had significantly (P≤0.05) lower levels of N, P, K, Ca and Mg, in fact, all below the RDA. The low levels could partly be attributed to the slightly acidic nature of Lela soils which may have hindered the uptake of K, Ca and Mg. In addition, the soil pH was below optimum range (5.5-7.5) for growing most ALVs [52]. Low soil pH in soils from Lela site may have resulted in low P bioavailability due to high P fixation capacity; toxicities of Fe and Mn and deficiencies of K, Ca, Mg and Zn [53]. These deficiencies or toxicities often act together to limit plant growth [54]. This may have limited the uptake of P, K, Ca, Mg and Zn and enhanced the uptake of Mn by ALVs grown in Lela site. Similarly, the high levels of Na and Mn in soils from Lela site could have inhibited the uptake of other nutrients due to the antagonism between Na and K which may have resulted in direct osmotic gradient [55]. This may have resulted in a high influx of Na into the root cells. Also, Na may have enhanced the efflux of K into the growth medium because of disturbed membrane integrity [56]. High Na levels in the external medium greatly reduces the activity of Ca in the solution and may result in a decrease in the amount of Ca available for uptake by the plants [57,58]. Calcium is strongly competitive with Mg. The binding sites on the root plasma membrane appear to have less affinity for the highly hydrated Mg than for Ca [25] which explains the higher intake of Ca compared to Mg.

Temperature usually contributes towards the crop growth, development, and productivity. The change from vegetative to reproductive phase is greatly influenced by temperature changes. The mean temperature of 25.7, 23.9, and 21.9ºC were reported in Busia, Kisumu, and Lela, respectively during the study period (Table11).

The air temperature was within the optimal range (20-30ºC) for the growth of most vegetables [30,59] in the three locations. Therefore, the variation in the levels of nutrients in ALVs cannot be attributed to variation in air temperature. The rainfall data during the study period is presented in (Table 12). Busia had the highest amount of rainfall while Lela had the least.

Table 9. Variation in levels of Na (mg/100 g DW) in selected ALVs at different location and harvesting stages

Loc	H. stage (wks)	Species					
		V. unguiculata	***A. Hybridus***	***C. gynandra***	***S. scabrum***	**M. stage**	**M. loc**
	4	25.621	195.150	13.483	64.047	74.621	
Busia	6	25.593	152.557	13.593	63.673	63.854	
	8	24.697	151.447	13.463	64.757	63.591	
	10	25.783	156.537	13.290	64.470	65.020	
	Mean species	25.470	163.923	13.457	64.237		66.772
	CV (%)			26.930			
	LSD (P≤0.05)			16.606		NS	
	Interaction			NS			
Kisumu	4	22.137	171.840	16.307	64.910	68.798	
	6	22.327	242.310	16.267	66.483	86.847	
	8	22.247	180.210	16.620	65.360	71.109	
	10	22.677	186.993	17.027	67.443	73.535	
	Mean species	22.347	195.338	16.049	66.049		75.072
	CV (%)			19.410			
	LSD (P≤0.05)			18.925		NS	
	Interaction			26.907			
Lela	4	41.630	187.707	22.713	86.603	84.663	
	6	43.193	187.990	23.143	88.313	85.660	
	8	42.787	192.603	22.920	88.417	86.682	
	10	42.973	195.037	23.353	87.683	87.262	
	Mean species	42.646	190.834	23.032	87.754		86.067
	CV (%)			1.220			
	LSD (P≤0.05)			1.359		1.359	
	Interaction			1.932			
all sites	4	29.857	184.899	17.501	71.853	76.028	
	6	30.371	194.286	17.668	72.823	78.787	
	8	29.910	174.753	17.668	72.844	73.794	
	10	30.478	179.522	17.890	73.199	75.272	
	Mean species	30.154	183.365	17.682	72.680		75.970
	RDA (mg)						2300.000
	AVC*						1.747*
	CV (%)			17.460			
	LSD (P≤0.05)			9.946		9.946	11.647
	Interaction			22.748			

*AVC *, average amount of vegetables (kg) to be consumed daily by a healthy adult to supply RDA*

Table 10. Initial soil pH and total nutrients N (%), P, K, Ca, Mg, Fe, Zn, Mn and Na in mg/100 g (DW) by locations

Nutrient/Location	Busia	Kisumu	Lela	CV%	LSD (P≤0.05)
pH	6.467	6.800	5.200	1.080	0.234
N	0.453	0.600	0.277	16.900	0.263
P	124.299	202.039	63.534	10.780	49.199
K	272.385	379.088	203.088	5.530	55.395
Ca	848.974	1071.407	568.219	4.610	134.221
Mg	467.841	728.285	354.631	3.060	55.600
Fe	1210.713	1033.021	639.859	5.300	178.832
Zn	7.565	9.941	4.965	6.370	1.677
Mn	0.234	0.167	0.453	20.480	0.205
Na	252.449	387.668	512.948	5.750	77.700

Table 11. Maximum, minimum and mean temperatures (ºC), April-September, 2009

Temp.	Location	Months						
		April	May	June	July	August	Sept	Mean.loc
Max. temp	Busia	38.5	38.3	38.3	38.0	30.0	38.0	36.9
Min. temp		14.0	14.0	11.7	11.0	13.0	12.0	12.6
Mean. temp		26.3	26.0	25.0	25.0	26.0	26.0	25.7
Max. temp	Kisumu	29.7	30.1	30.4	30.0	30.8	31.9	30.5
Min. temp		18.2	17.9	16.4	16.1	17.4	17.8	17.3
Mean. temp		24.0	24.0	23.4	23.0	24.3	24.9	23.9
Max. temp	Lela	29.1	30.5	29.8	28.7	29.9	30.0	29.7
Min. temp		13.7	14.0	13.6	13.5	13.8	14.5	13.9
Mean. temp		21.4	22.3	21.7	21.1	21.9	22.3	21.9

Source: KARI-Alupe, KARI-Kisumu, and ATC-Maseno

Table 12. Total and mean rainfall (mm) received (April- September 2009)

	Location	Months						
		April	May	June	July	August	Sept	Mean.loc
Total	Busia	326.7	268.2	48.9	95.4	86.7	214.1	
Mean		11.0	17.9	5.4	23.9	6.2	15.3	13.3
Total	Kisumu	270.4	124.2	53.1	101.7	89.3	229.6	
Mean		12.3	7.8	5.9	12.7	7.4	14.4	10.1
Total	Lela	198.7	152.5	137.8	41.4	97.5	175.5	
Mean		18.1	19.0	19.6	8.3	12.1	14.6	15.3

Source: KARI, Alupe; KARI, Kibos and ATC, Maseno

During the study period, over half the annual precipitations of 1040.0, 868.3, and 803.4 mm were received in Busia, Kisumu, and Lela, respectively. April-May-June, 2009 showed deficient rainfall of 643.8, 447.7, and 489.07

3.2 Effect of Harvesting Stage on Levels of Nutrients in Selected ALVs

The levels of N, P, K, Ca, and Zn in ALVs significantly (P≤0.05) increased from 4 to 6 WAE then decreased from 6 to 10 WAE, Fe levels increased (from 4 to 6 WAE) while Mg levels increased (from 4 to 8 WAE) then decreased from 8 to 10 WAE. The Na and Mn levels did not vary significantly with harvesting stage. Generally, optimum levels of N, P, K, Ca, Mg, Fe and Zn were reported at 6 WAE. Thereafter, the levels of nutrients significantly decreased. Variations in the levels of nutrients in ALVs at different harvesting stages had been reported in Nairobi, Kenya [34] Zimbabwe [35] and South Africa [30]. Highest levels of Ca and Zn in *Amaranthus cruentus* were attained at 3 WAE, K at 6 WAE and P at 4 WAE in 489.0 mm and 396.2, 420.6, and 314.4 mm in July-August-September in Busia, Kisumu, and Lela, respectively. The rainfall in the three locations was within the optimal range required for vegetable growing [52].

Zimbabwe [35] and South Africa [30]. However, highest levels of Na were reported at 6 WAE in Zimbabwe [35] and 7 WAE in South Africa [30]. The levels of N significantly (P≤0.05) increased from 4 to 6 WAE contrary to the results reported in spider plant in Nairobi, Kenya [34]. Differences at the stage at which highest levels of nutrients were attained in the current study and the previous studies could be attributed to variations in the initial soil nutrient levels, agronomic practices, and environmental conditions of the study sites.

The uptake rate of many nutrients depends on the nutrients demand for growth [60,61] determined by the role the nutrients play in plant growth. Most of the functions of Ca and P are as structural components of macromolecules, K is required for cell expansion, protein synthesis and in stomatal regulation, while Mg is the central atom in the chlorophyll molecule. The uptake of N, P, K, Ca, and Mg was high during the vegetative stage when roots were actively growing than in reproductive stages [62]. This explains the significant increase in the levels of N, P, K, Ca, Mg, and Zn from 4 to 6 WAE. During the reproductive stage, nutrients are partitioned towards the reproductive organs [63]. In addition, translocation of nutrients N, P and K in the form of amino acids from shoots to roots in the absence of soil nutrient replenishment and a decrease in demand for nutrients for new growth as plants age accounts for the decrease in the levels of nutrients in ALVs with increasing plant age. The results of this study are in agreement with findings of Marschner [64].

The non-significant increase in levels of Na and Mn in ALVs with increasing plant age may partly be attributed to the role these nutrients play in plant growth. Manganese plays an important role in redox processes and is required in very small amounts while Na is a beneficial element required for osmotic adjustment [65]. Its uptake is concentration dependent with no specific binding sites in the plasma membrane [66].

There was significant ($P \le 0.05$) interaction between species, harvesting stage and location of production for all the nutrients except K, Ca and Fe suggesting the response patterns were different at each harvesting stage and location for each species. The ALVs from the three locations are significant contributors of Mg and Fe and non-significant contributors of P, K, Mn and Na while ALVs from Kisumu site are significant contributors of Ca and Zn. When vegetables are bought from the market, their source is unknown, therefore, on average, consumption of 3.577, 3.608, 0.506, 0.270, 0.199, 0.588, 3.529 and 1.747 kg DW of the four species is recommended to supply RDAs of P, K, Ca, Mg, Fe, Zn, Mn and Na, respectively.

4. DISCUSSION AND CONCLUSION

From this particular study, it has been observed that, amongst *Vigna unguiculata*, *Amaranthus hybridus*, *Cleome gynandra* and *Solanum scabrum vegetables only Amaranthus hybridus* had significantly ($P \le 0.05$) higher levels of P, Ca, Zn, Mn, and Na. The potential of generating several macro and micronutrients in food is much higher in it. All the nutrients increase its level at first then with longer duration of farming it starts to detoriate its value. The levels of nutrients significantly ($P \le 0.05$) varied with different site except the nutrient level of Na, which remains constant. The ALVs from Kisumu site had significantly ($P \le 0.05$) higher levels of N, P, K, Ca, Mg and Zn, ALVs from Lela site had higher levels of Mn and Na while ALVs from Busia site had higher levels of Fe.

On the contrary it bcan nbe said that *Amaranthus hybridus* is a better contributor of-of P, Ca, Zn, Mn, and Na. The Fe, Mg and Zn levels were above the Recommended Dietary Allowance (RDA) and may be used to alleviate deficiencies associated with such nutrients. Healthy farming techniques are needed to be involved to increase the proposed nutritional values of the food. There is a need for the development of regional food composition tables for all ALVs in Kenya.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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