

Effect of Different Growing Periods on the Nutritional Composition of Cowpea (*Vigna unguiculata*) Leaves

ABSTRACT

Introduction: The nutritional composition of plant tissues is influenced by climatic conditions during their growth and development. Climate influences the growth rate, chemical composition and yield of crops. This study was conducted to evaluate the effect of growing periods on the nutritional composition of six cowpea varieties; Kenyan local variety (KLV), Indonesian local variety (ILV), Malaysian local variety (MLV), Japanese local variety (JLV) and two Kenyan varieties (K80 and KenKunde).

Methodology: The seeds were grown in an experimental glasshouse under three varying growing periods in 2015 and 2016, respectively. The effect of growing condition on moisture, protein, potassium, calcium, magnesium, manganese, iron and zinc content was assessed.

Results: Moisture content varied from 67.9% to 78.0% and 81.1 % to 88.8% in 2015 and 2016, respectively; while protein contents ranged from 7.3% to 14.6% and 18.4% to 30.4% in 2015 and 2016, respectively. In 2015, there was a significant increase in moisture content, protein, K, Ca, Mg and Mn in KLV as temperature increased. This trend was different for ILV and MLV, which showed significant differences across growing periods with the June growing period showing the highest moisture, protein, K, Mg and Mn contents. In 2016, protein, Ca and Mg contents increased in response to increasing temperature across the growing periods. KLV and KenKunde exhibited higher concentrations of most components.

Conclusion: There were significant differences among varieties and across growing periods in nutrient content implying both variety and climatic conditions affect nutritional composition.

Keywords: Cowpea leaves, nutritional composition, growing period

1. INTRODUCTION

Cowpea (*Vigna unguiculata*) is an annual herbaceous warm season leguminous vegetable believed to be one of the oldest crops to be farmed in Africa where it was first domesticated (OECD, 2016; Timko & Singh, 2008). Further domestication probably occurred in Asia, before spreading into Europe and America (Xiong et al., 2016). Cowpea plays an important role in the nutritional security of many people in semi-arid regions of developing countries with the main product being the dry grain (OECD, 2016). However, utilization varies among and within regions; for instance in many parts of East Africa, leaves are often consumed either cooked by boiling or fried and eaten as an accompaniment to a stiff porridge (*Ugali*) besides drying and milling into powder for later use when they are out of season (Ahenkora, Adu Dapaah, & Agyemang, 1998). Green peas are consumed in the southern United States

and Senegal, fresh green pods are consumed in the humid regions of Asia, while the whole plant is also used as green manure in southern United States and Australia and as fodder in parts of the Sahel (Ehlers & Hall, 1998).

In Africa, leafy cowpea is classified and referred to as an African indigenous vegetable (AIV) (Ekesa, Walingo, & Onyango, 2009; Okello et al., 2015). Like other AIVs, it requires low levels of investment and produce high yields hence play an important role in the livelihoods of the rural and urban dwellers by providing necessary nutrition, employment and is a source of income especially for women who are involved in their production (Muhanji, Roothaert, Webo, & Stanley, 2011). Cowpea for leaf production has a short growing period of 21-42 days, while cowpea for seed production requires a longer period of 70-120 days (Ohler, Nielsen, & Mitchell, 1996), therefore leafy cowpea can be grown and consumed throughout the year. Cowpea has considerable ability to adapt to high temperatures and drought as compared to other food crop species (Ehlers & Hall, 1998) and while it is mainly grown in hot low elevation equatorial and subtropical areas, in Kenya, it can be grown at altitudes of up to 1600m above the sea level. In addition to its ability to fix nitrogen, cowpea also possesses an extensive root system that helps prevent soil erosion besides the ability to withstand both acidic and alkaline soil conditions (Gomez, 2004).

Cowpea leaves have many health benefits including a good source of vitamins, mineral salts and antioxidants, and like other greens, the leaves have high fiber content (Drahansky et al., 2016; Khalid & Elhardallou, 2016). A diet containing this vegetable is great for diabetic, cardiovascular and overweight conditions. The minerals in cowpea leaves are known to be more bioavailable than those in its seeds due to the presence of phytic acid in the seeds which reduces the bioavailability of minerals such as calcium and iron (Khalid & Elhardallou, 2016; Sinha & Kawatra, 2003). Moreover, the protein content in the leaves is higher ranging between 29-41% as compared to that in seeds which is 21-33% (Ahenkora et al., 1998). Dry grain production is the only commodity of cowpea formerly estimated on a worldwide basis. For instance, the Food and Agricultural Organization (FAO) estimate that nearly 4 million metric tons is produced annually on about 10 million ha while a 1997 estimate suggests that cowpeas are cultivated on 12.5 million hectares with a worldwide production of 3 million tons and are consumed by 200 million people on a daily basis (Gomez, 2004). Most cowpeas are grown on the African continent, particularly in Nigeria and Niger which account for 66% of world production.

The nutritional composition and quality of fruits and vegetables can be influenced by various factors such as genotypic differences, pre-harvest climatic conditions and cultural practices, maturity and harvesting methods, and postharvest handling procedures. For instance, climatic conditions including light and average temperature which vary considerably depending on the season, strongly influence the chemical composition of horticultural crops (Hornick, 1992). Thus, the locations and seasons in which these plants are grown are likely to determine the contents of functional components such as ascorbic acid, carotene, riboflavin, and thiamine, among others; with low ascorbic acid in plant tissues attributed to low light intensity (Verkerke, Labrie, & Dueck, 2015). Rainfall also determines the amount of water supplied to the plant which may in turn affect the composition of the harvested product and its susceptibility to mechanical damage and decay during subsequent harvesting and handling operations. Temperature on the other hand influences the nutritional composition during growth and development; as temperature increases up to a point, photosynthesis, transpiration, and respiration increase. High temperatures increase respiration, sometimes above the rate of photosynthesis, thus, photosynthates are used up faster than they are produced. For optimum growth, photosynthesis must be greater than respiration (Lee & Kader, 2000).

Most of the cowpea research has mainly focused on the production of seeds with little research on cowpea as a leafy vegetable being undertaken hence limiting the exploitation of this important food source. Furthermore, most nutrient database available provide only year-round averages for nutritional content which may not be representative of the variability that

is dependent on different varieties, management practices, seasonality, stage of harvest, environment, soil among other factors (Rickman, Bruhn, & Barrett, 2007). While cowpea is regarded as a drought resistant crop, variation in nutrient contents has been reported in several varieties investigated (Ahenkora et al., 1998). Variation in protein content in particular has been reported in cowpea varieties grown in different seasons (Sebetha, Ayodele, Kutu, & Mariga, 2010). Therefore this study was conducted to investigate the effect of different growing periods on the nutritional composition of cowpea leaves in order to optimize its potential contribution to nutrition and health.

2. MATERIAL AND METHODS

2.1 Cowpea varieties

This study used six cowpea varieties; KLV (Kenyan variety M66), ILV (Indonesian local variety) and MLV (Malaysian local variety) were used in 2015, while in 2016 the varieties used were K80, KenKunde (Kenyan varieties) and JLV (Japanese variety).

2.2 Experimental design and planting

The varieties were grown in a glasshouse in Kyoto University under similar conditions. Sowing was done in three consecutive periods; May, June and July.

A randomized complete block design (RCBD) was employed with three replications for each variety. Seeds of each variety were sown in small plastic pots using a soil mixture of commercial planting soil and sand at the ratio of 1:1. Watering was done daily in the evening.

Leaves were harvested 6 weeks after planting with harvest done in early morning. The harvested leaves were put in clear zip lock bags and stored in a cool box until analyses.

2.3 Nutritional analysis

2.3.1 Proximate analysis

The moisture content was determined by weighing fresh leaves using an analytical weighing balance, after which they were oven dried for 2 days at 70°C using a constant temperature oven (Model DN-41, Yamato scientific Co., Ltd, Tokyo, Japan). The dry weight was measured and percentage moisture content calculated. The dried sample was analyzed for protein content following AOAC methods (AOAC, 2000). The crude protein content was determined from combustion of approximately 20mg of the ground sample by an automatic high sensitive nitrogen carbon analyzer (NC-22F, Sumika Chemical Analysis Services, Ltd, Tokyo, Japan). Total crude protein level was obtained by multiplying the total N value by the conversion factor of 6.25 and expressed in percentage (AOAC method 955.04). All the analyses were carried out in triplicate.

2.3.2 Determination of mineral contents

The dried leaves were ground using a ball mill (Yoshida Seisakusho Co. LTD, Tokyo, Japan). The ground samples were placed in airtight containers and stored in a desiccator until used for the analyses. Each sample (0.1g) was weighed in duplicate using an analytical weighing balance and put in test-tubes. 2ml of concentrated nitric acid and 0.2ml of perchloric acid were added and the samples digested in a block digester (Model MB.3H.U, Koike Precision Instruments, Japan). After complete digestion, the samples were dissolved in 0.1% nitric acid to adjust the volume to 20ml. 250µl of the sample was diluted with 5ml of 0.1% nitric acid for potassium determination. 200 ml of sample was diluted with 5ml of 0.1% nitric acid for calcium and magnesium determination. 52µl of lanthanum chloride solution was then added to avoid interference of other elements in the sample. For Mn, Zn and Fe determination, 5ml of the original sample was used. The contents were mixed well using a vibrator test tube mixer (Pasolina NS-80). The samples in the test-tubes were placed in an auto sampler (ASC-6100, Shimadzu Corporation, Tokyo, Japan) and determined using an atomic absorption flame emission spectrophotometer (AA-6200, Shimadzu Corporation, Tokyo, Japan). The analyses were carried out in triplicate and the minerals were expressed in mg/g dry weight.

2.4 Data analysis

All experiments were carried out in triplicates unless stated otherwise, and the mean value \pm standard deviation (SD) was used in data analysis. The Analysis of variance, means, standard deviation and Tukey- Kramer honest significant difference (HSD) test following analysis of variance were performed using JMP (SAS Version 13).

3. RESULTS AND DISCUSSION

The interactions between the effects of growing periods and varieties found in some parameters in each combination of growing periods and varieties are shown for all the parameters measured.

3.1 Temperature during growing periods

Air temperatures during the growing periods of 2015 are shown in Figure 1. Mean temperature during May growing period was 24°C with a minimum of 11.9°C and a maximum of 38.4°C. Mean temperature during June growing period was 25.9°C with a minimum of 17.6°C and a maximum 39.4°C. In July growing period, mean temperature was 27.7°C with a minimum temperature of 20.6°C and a maximum of 42°C.

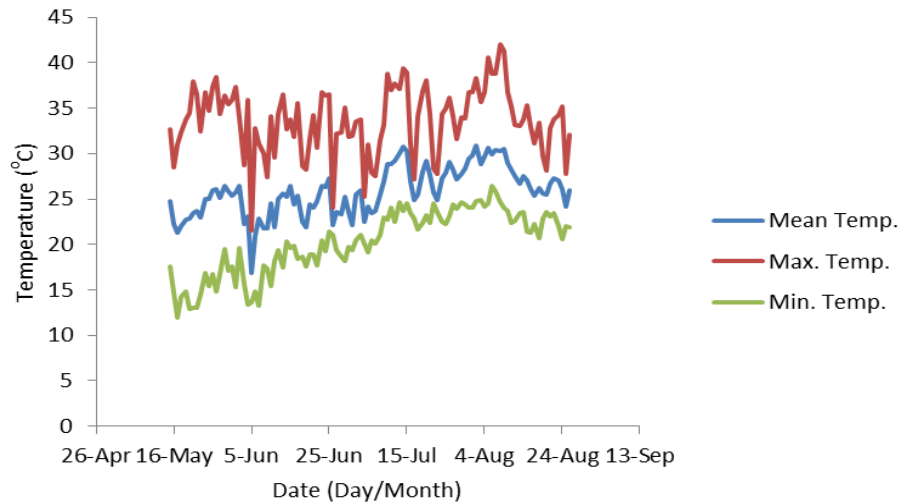


Figure 1. Changes of mean, maximum and minimum temperatures during the year 2015 experimental period.

Air temperatures during the growing periods of the year 2016 are shown in Figure 2. Mean temperature during May growing period was 23.5°C with a minimum of 9.6°C and a maximum of 36.0°C. In June growing period, mean temperature was 25.7°C with a minimum of 16.7°C and a maximum of 34°C while in July growing period, the mean temperature was 28.2°C with a minimum of 19.4°C and a maximum of 40.3°C.

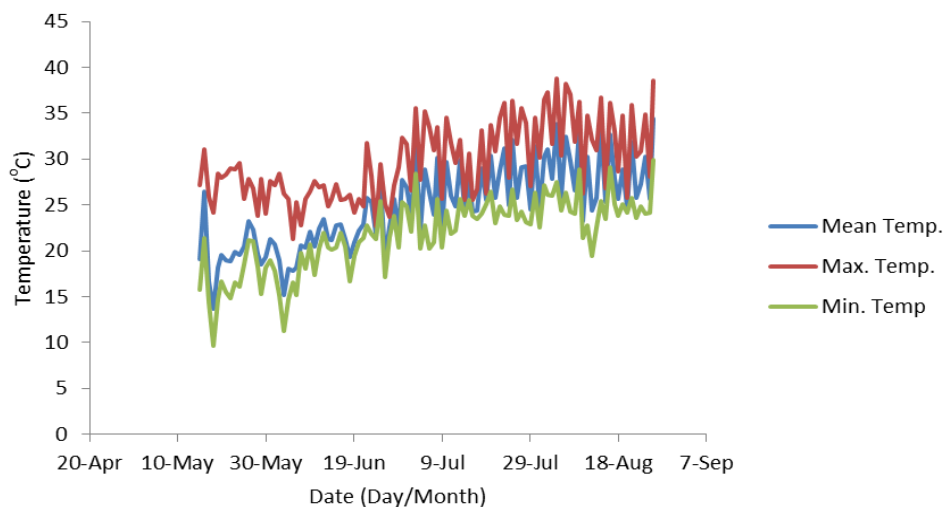


Figure 2. Changes of mean, maximum and minimum temperatures during 2016 experimental period

3.2 Moisture and protein composition of cowpea varieties in 2015 and 2016

Moisture contents were significantly affected by the growing period. The month of May growing period exhibited lower moisture contents than June and July in all the varieties (Table 1). There were also significant differences between varieties with KLV showing higher moisture contents compared to ILV and the MLV.

Protein contents were also influenced by the growing period with June generally showing the highest protein contents (Table 1). However, the protein contents differed significantly between KLV and the other two varieties with KLV showing higher amount of protein in all the growing periods (Table 1).

Table 1. Moisture contents and protein compositions of leaves of cowpea varieties in 2015

Component (%)	Variety	Growing period		
		May	June	July
Moisture	KLV	77.95±1.68aB	84.11±1.07aA	85.36±1.85aA
	ILV	68.65±1.46bB	76.36±1.37bA	74.68±2.01bA
	MLV	67.89±0.66bC	75.53±1.03bA	73.62±1.81bB
Protein	KLV	10.14±0.73aB	13.31±1.72aA	14.57±2.23aA
	ILV	7.55±0.58bB	9.50±1.00bA	7.31±0.88bB
	MLV	7.85±0.34bB	9.61±1.11bA	7.95±0.84bB

Values followed by different lower case letters within a column and different upper case letters within a row indicate a significant difference (Tukey's test).

P=.05, n=6).

Moisture contents were significantly different across the growing periods (Table 2) with June growing period exhibiting higher moisture contents for all the varieties. There were no significant differences in moisture contents among varieties in July. In May growing period, JLV showed lower moisture content, whereas in June growing period, Kenkude showed higher moisture content. The maximum water content may have varied due to the different growing periods that could have influenced structural differentiation.

Protein contents were influenced by the growing period with June and July growing periods exhibiting higher protein contents compared to May growing period (Table 2). Protein contents differed significantly between JLV and the other two varieties with JLV showing lower protein contents in all the growing periods.

Although previous studies have reported that plants exposed to cool temperatures perform in a similar way as plants exposed to drought conditions in terms of water content (Rodríguez et al., 2015) this experiment was conducted under adequate irrigation to eliminate any effect of drought conditions on the physiological response. This could explain why there was no reduction of moisture contents in the July growing period, which had high temperature with a mean of 27.7°C in 2015 and 28.2°C in 2016. In actual cultivation conditions in the tropics, however, farmers frequently face the problem of drought. Further studies are necessary to seek for varieties that can maintain relatively high moisture contents under drought conditions.

Table 2. Moisture and protein composition of leaves of cowpea varieties in 2016

Component (%)	Variety	Growing period		
		May	June	July
Moisture	K80	85.57±1.83aB	87.68±0.70bA	84.13±0.82aB
	Kenkude	84.99±1.36aB	88.79±0.29aA	85.19±1.60aB
	JLV	81.08±1.90bC	86.99±0.85bA	83.77±0.80aB
Protein	K80	24.58±3.28aB	27.47±1.39aAB	28.72±1.29abA
	KenKude	22.91±3.80abB	29.77±1.80aA	30.44±1.48aA
	JLV	18.37±3.11bB	24.77±2.00bA	27.02±1.33bA

Values followed by different lower case letters within a column and different upper case letters within a row indicate a significance difference (Tukey's test,

$P=0.05$, $n=6$).

Protein contents ranged from 7.3% to 14.6% in 2015 and from 18.4% to 30.4% in 2016 (Table 1 and 2). The results in 2016 were consistent with ranges (23.9%-30.9%) reported in a previous study (Ddamulira, Fernandes Santos, Obuo, Alanyo, & Lwanga, 2015). However, the protein contents for varieties grown in 2015 were very low suggesting that the leaves of some cowpea varieties may have comparatively lower protein contents than those that have been previously reported. Protein contents can be as high as 36% dependent on genotype and environmental factors (Dakora & Belane, 2019; Gonçalves et al., 2016; Sebetha, Modi, & Owoeye, 2014). The protein contents for the three varieties in 2016 were higher than the contents reported for *Momordica balsamina* (11.29%) and *Moringa oleifera* (20.72%), which are considered as high value leafy vegetables (Aberoumand & Deokule, 2009). The contents were also comparable to that of Spinach (28%), but higher than those of cabbage

(15.4%) (Sebetha et al., 2010). Plants that contribute more than 12% of their calorific value from proteins are known to be good sources of protein (Ehilé, Kouassi, Kouamé, Denis Yao, & Amani, 2018). Several studies have indicated that African indigenous vegetables may contain higher nutrient levels than those found in exotic vegetables (Steyn, Olivier, Winter, Burger, & Nesamvuni, 2001) hence could provide a readily available source of cheap protein for improved nutrition. While African cowpea varieties in this study showed higher protein contents than Asian varieties, the number of varieties used was limited. The African varieties, developed for use as leafy vegetables may have higher protein contents than Asian varieties, in which breeding for use as leafy vegetables has not been done.

Higher temperatures lead to higher leaf protein turn-over in *Amaranthus* spp.; hence the protein contents are lower under high temperature conditions than under cool conditions (Modi, 2007). This is attributed to increased maintenance respiration, which occurs at the expense of growth respiration under high temperature conditions. However, in this study the protein contents increased with increasing temperature across the growing periods to some extent. This may be related to a general trait of cowpea, which is believed to be well adapted to high temperature conditions. In early growing season, low mean and minimum temperatures in several days may have affected protein biosynthesis adversely. In addition, cowpea is known as a heat tolerant crop, and the reduction of protein synthesis may not have been observed.

3.3 Mineral composition of cowpea leaves

Potassium contents were significantly affected by the growing period. The June growing period showed higher potassium contents in all the varieties (Table 3) while there were no significant differences between varieties except for KLV in May growing period, which showed lower potassium contents compared to the ILV and MLV.

Calcium contents, on the other hand, increased with increasing temperatures across the growing periods (Table 3). The May growing period showed lower calcium contents compared to the other growing periods. Moreover, there were significant differences between varieties with KLV showing higher calcium contents in all the growing periods (Table 3). On the other hand Magnesium contents were higher in June growing period in ILV and MLV, while July growing period showed lower contents of Mg in those varieties (Table 3). However, in KLV there was a significant increase from May growing period to June and July growing period. KLV generally showed higher Mg contents than the other 2 varieties.

Manganese contents also showed significant differences across the growing periods with June growing period showing higher content in ILV and MLV (Table 3). In KLV, June and July growing periods showed higher Mn contents than May growing period. There were no significant differences between varieties for the growing periods of May and July. In June growing period, KLV showed lower Mn contents than MLV.

Iron content was not significantly affected by the growing period in all the varieties and the differences between varieties were not significant in June and July growing periods (Table 3). In May growing period, MLV showed higher iron content followed by ILV, whereas KLV had the lowest iron content.

Zinc content in KLV and ILV was not significantly affected by the growing period but the levels in MLV were significantly higher in June than in May and July growing periods (Table 3). There were no significant differences in Zn contents between varieties in all the growing periods.

Table 3. Mineral composition of leaves of cowpea varieties in 2015

Component (mg/g)	Variety	Growing period		
		May	June	July
K	KLV	8.59±0.43bB	12.67±1.66aA	12.01±1.85aA
	ILV	9.79±0.68aB	12.91±1.42aA	11.09±0.67aB
	MLV	9.95±0.59aB	14.87±1.29aA	11.75±1.97aB
Ca	KLV	10.19±1.05aC	14.72±0.87aB	17.70±2.30aA
	ILV	5.49±0.48cC	8.91±0.58cB	10.0±0.64bA
	MLV	6.83±0.92bB	10.45±1.03bA	9.69±0.47bA
Mg	KLV	2.14±0.22aB	3.27±0.31aA	3.48±0.50aA
	ILV	1.88±0.12bB	2.22±0.26bA	1.73±0.07bC
	MLV	1.92±0.08abB	2.29±0.27bA	1.59±0.07bC
Mn	KLV	0.34±0.04aB	0.44±0.06bA	0.44±0.08aA
	ILV	0.30±0.03aC	0.50±0.05abA	0.37±0.04aB
	MLV	0.32±0.03aC	0.55±0.03aA	0.37±0.03aB
Fe	KLV	0.05±0.01cA	0.10±0.02aA	0.16±0.22aA
	ILV	0.07±0.02bA	0.11±0.05aA	0.14±0.08aA
	MLV	0.10±0.02aA	0.15±0.04aA	0.10±0.06aA
Zn	KLV	0.003±0.004aA	0.002±0.002aA	0.005±0.003aA
	ILV	0.004±0.002aA	0.003±0.001aA	0.005±0.001aA
	MLV	0.003±0.001aB	0.001±0.002aB	0.006±0.001aA

Values followed by different lower case letters within a column and different upper case letters within a row indicate a significance difference (Tukey's test,

P = 0.05, *n* = 6). Values are on dry matter basis.

Potassium content was significantly affected by the growing period with June and July showing higher contents than May growing period in Kenkunde and JLV, whereas in K80 June growing period showed higher K content than May growing period (Table 4). However, there were no significant differences among the varieties in all the growing periods.

Potassium is known to have a bigger presence in both fruits and vegetables, however nitrogen and calcium show major impacts on horticultural crop quality. Potassium is the most abundant individual mineral element in fruits and vegetables. It normally varies between 60 and 600 mg per 100 g⁻¹ of fresh tissue (Wojciech et al., 2009) which is consistent with the figures obtained in this study.

Calcium and Magnesium contents also increased across the growing periods with June and July growing period showing higher contents in all the varieties (Table 4). JLV showed the lowest calcium content while KenKunde had the highest magnesium content among varieties used in all the growing periods. Calcium is believed to have a major influence on the rheological properties of the cell wall and, consequently, on the texture and storage life of fruits and vegetables. Magnesium is important in protein synthesis, release of energy from muscle storage and body temperature regulation. Generally, magnesium levels are significantly higher in vegetables than in fruits (Wojciech et al., 2009)

Table 4. Mineral composition of leaves of cowpea varieties in 2015

Component (mg/g)	Variety	Growing period		
		May	June	July
K	K80	20.92±2.84aB	28.96±3.18aA	24.85±2.74aAB
	KenKunde	21.81±2.19aB	31.25±2.69aA	27.81±5.47aA
	JLV	18.71±3.02aB	27.64±1.63aA	24.41±2.22aA
Ca	K80	15.77±1.25aB	21.23±1.84aA	22.60±1.69aA
	KenKunde	14.32±0.88abB	21.13±1.79aA	20.90±3.06abA
	JLV	12.68±2.24bB	17.72±0.72bA	19.35±1.34bA
Mg	K80	2.23±0.21bB	3.26±0.17bA	3.57±0.32bA
	KenKunde	2.75±0.29aB	3.92±0.18aA	4.23±0.54aA
	JLV	1.93±0.38bB	2.95±0.35bA	3.14±0.28bA
Mn	K80	0.55±0.05bB	0.82±0.10aA	0.59±0.08bB
	KenKunde	0.73±0.02aB	0.96±0.16aA	0.82±0.08aAB
	JLV	0.72±0.07aA	0.82±0.05aA	0.82±0.10aA
Fe	K80	0.22±0.05aA	0.18±0.05aA	0.20±0.08aA
	KenKunde	0.20±0.08aA	0.17±0.04aA	0.21±0.07aA
	JLV	0.22±0.04aA	0.13±0.04aB	0.21±0.06aA
Zn	K80	0.028±0.007aAB	0.036±0.006aA	0.02±0.004bB

KenKunde	0.035±0.004aA	0.04±0.004aA	0.03±0.01aA
JLV	0.033±0.007aA	0.04±0.004aA	0.025±0.004abA

Values followed by different lower case letters within a column and different upper case letters within a row indicate a significance difference (Tukey's test).

$P=0.05$, $n=6$). Values are on dry matter basis.

Manganese content was significantly affected by the growing period with June growing period showing higher contents in K80 and KenKunde (Table 4). However, there was no significant difference in manganese content in JLV in all the growing periods. K80 showed lower Mn content than the other varieties in May and July growing periods. In plants, manganese atoms appear to undergo successive oxidations to yield a strongly oxidizing complex that is capable of water oxidations during photosynthesis. Similarly like magnesium, manganese is required in enzyme reactions involving carbon assimilation.

Iron content was not significantly affected by the growing period in K80 and KenKunde (Table 4) but was lower for JLV in June than those in the other growing periods. There was no significant difference in iron content among varieties in all the growing periods.

Iron is a constituent of the haem complex, a naturally occurring plant chelate involved in electron transfer in a number of important plant enzymes. In comparison to vitamins, minerals are more stable and their contents are not affected significantly by cooking. Cooking Leafy vegetables in iron utensils increases the total iron and bioavailable iron compared to fresh vegetables besides those cooked in other metallic utensils such as stainless steel and aluminum a phenomenon that could also apply to ALVs (Nangula et al., 2010).

Zinc contents in KenKunde and JLV were not influenced by the growing period (Table 4), whereas the content was lower for K80 in July than in June growing period. There was no significant difference in Zn content among varieties for May and June growing periods while in July growing period, Zn content was lower in K80 than in KenKunde. Zinc plays a catalytic or a structural role in more than 200 enzymes involved in digestion, metabolism, reproduction, and wound healing. In addition, zinc has a critical role in immune response, and is an important antioxidant.

Mineral contents in 2015 ranged from 8.6 to 14.8 mg/g for potassium, 5.5 to 17.7 mg/g for calcium, 1.6 to 3.48 mg/g for magnesium and 0.3 to 0.6 mg/g for manganese, while in 2016 they ranged from 18.7 to 31.3 mg/g for potassium, 12.7 to 21.2 mg/g for calcium, 1.9 to 4.2 mg/g for magnesium and 0.6 to 1.0 mg/g for manganese. The results also indicate iron levels of up to 0.2 mg/g and Zinc contents of 0.1 mg/g (Table 3 and 4). These values are comparable to values obtained from indigenous leafy vegetables investigated previously (Nyadanu & Lowor, 2014; Onyango, Imungi, & Harbinson, 2008).

The structure of leaf canopy and position of leaf tissue influences the leaf nutrient content (Sebetha et al., 2010). This could be one of the reasons behind the varietal differences in nutrient composition since the varieties have different leaf structures. Moreover, the nutrient content of raw plant foods vary widely and is affected by factors such as variety or cultivar; part of the plant consumed; stage of maturity; geographic site of production or climate; harvesting and post-harvest handling conditions; and storage and thus comparing the nutrient content of leaves from different data sources is quite challenging (Jaarsvelaet al., 2014)

3.4 Correlations between moisture and nutritional components

The correlations between moisture contents and nutritional components are shown in Table 5. There were significant ($P=0.05$) correlations between moisture contents and protein, calcium and manganese in 2015 and potassium in 2016.

Table 5. Correlation coefficients between moisture contents and nutritional components

Year		Correlation coefficients						
		Protein	K	Ca	Mg	Mn	Fe	Zn
2015	Moisture	0.92*	0.36	0.95*	0.86	0.51*	0.40	-0.07
2016	Moisture	0.60	0.83*	0.52	0.51	0.50	-0.60	0.44

*Significant at $P=0.05$

3.5 Correlations between Temperature and nutritional components

The correlations between temperature and nutritional components are shown in Table 6. Temperature did not show any significant ($P=0.05$) correlation with all the nutritional components.

Table 6. Correlation coefficients between temperature and nutritional components

	Correlation coefficients							
	Moisture	Protein	K	Ca	Mg	Mn	Fe	Zn
Temperature	0.17	0.17	0.18	0.43	0.44	0.12	0.13	-0.02

The composition of plant tissues is influenced by temperature during their growth and development with the extent of high and low temperatures and the total available heat determining the growth rate, chemical composition and consequent yield of most horticultural crops (Bonhomme, 2000; Lee & Kader, 2000). Growth intensity is enhanced by a rise in temperature; however, a rapid decrease in growth begins once the optimum temperature is exceeded (Pietruszka, Lewicka, & Pazurkiewicz-Kocot, 2007). Temperature above the optimum also reduces the translocation of assimilates to the harvestable portion. Studies have shown that temperatures of 20 and 26°C increase vegetative growth in carrot (Abdel, 2016). This could explain high amounts of most components in June growing period, which had a mean of 25.9°C and 25.7°C in 2015 and 2016 respectively.

It is important to understand plants response to various environmental conditions since it is the basis for adopting technologies that will ensure high quality crop production. This understanding allows for manipulation of the plants and the environmental factors. For instance, environmental factors can be manipulated through greenhouse production, thus ensuring production of crops out of their growing season leading to constant production (Hewett, 2006). Consequently, the issue of seasonality of production, localization of production areas and quality of produce could be solved. Further studies are necessary to address these issues.

4. CONCLUSION

Cowpea leaves are a good source of nutrients hence their consumption should be encouraged among populations to address the issue of malnutrition. While cowpeas can

withstand unfavorable weather conditions, this study suggests that much variation occurs within and across varieties in terms of nutritional quality. Some varieties adapt better to varying growing conditions than others with Kenyan varieties found to have higher amounts of most of the nutritional constituents compared to Asian varieties. Similarly, genotype influences the nutritional composition of cowpea leaves and interacts with the growing period. Appropriate environmental conditions are therefore necessary for the production of leafy cowpea with optimum nutritional quality.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors

ETHICAL APPROVAL (NOT APPLICABLE)

REFERENCES

- Abdel, C. G. (2016). Photosynthesis of Four Radish (*Raphanus Sativus* L. Var. *Sativus*) Cultivars Grown in Controlled Cabinets under Varying Temperatures and Irrigation Levels = عملها السيطرة محكمة كاربينات في نميت الفجل من أ صناف لأربعة الضوئي التركيب . *Jordan Journal of Agricultural Sciences*, 12(2), 591–617. <https://doi.org/10.12816/0030040>
- Aberoumand, A., & Deokule, S. S. (2009). High Nutritional Value of non-conventional Balsam Apple (*Momordica dioica* Roxb .): A wild fruit vegetable. *International Journal of Applied Agricultural Research*, 4(3), 215–222.
- Ahenkora, K., Adu Dapaah, H. K., & Agyemang, A. (1998). Selected nutritional components and sensory attributes of cowpea (*Vigna unguiculata* [L.] Walp) leaves. *Plant Foods for Human Nutrition*, 52(3), 221–229. <https://doi.org/10.1023/A:1008019113245>
- AOAC. (2000). *AOAC Official Methods of Analysis* (17th ed.).
- Bonhomme, R. (2000). Bases and limits to using “degree.day” units. *European Journal of Agronomy*, 13(1), 1–10. [https://doi.org/10.1016/S1161-0301\(00\)00058-7](https://doi.org/10.1016/S1161-0301(00)00058-7)
- Dakora, F. D., & Belane, A. K. (2019). Evaluation of Protein and Micronutrient Levels in Edible Cowpea (*Vigna Unguiculata* L. Walp.) Leaves and Seeds. *Frontiers in Sustainable Food Systems*, 3(September 2019), 1–10. <https://doi.org/10.3389/fsufs.2019.00070>
- Ddamulira, G., Fernandes Santos, C. A., Obuo, P., Alanyo, M., & Lwanga, C. K. (2015). Grain Yield and Protein Content of Brazilian Cowpea Genotypes under Diverse Ugandan Environments. *American Journal of Plant Sciences*, 06(13), 2074–2084. <https://doi.org/10.4236/ajps.2015.613208>
- Drahansky, M., Paridah, M. ., Moradbak, A., Mohamed, A. ., Owolabi, F. abdulwahab taiwo, Asniza, M., & Abdul Khalid, S. H. . (2016). We are IntechOpen , the world ’ s leading publisher of Open Access books Built by scientists , for scientists TOP 1 % . *Intech*, i(tourism), 13. <https://doi.org/http://dx.doi.org/10.5772/57353>
- Ehilé, S. J. E., Kouassi, N. K., Kouamé, C. A., Denis Yao, N., & Amani, G. N. guessa. (2018). Nutritional composition of five spontaneous wild plants used as human foods in Côte d’ivoire areas (West Africa), a potential role in household food security. *Pakistan Journal of Nutrition*, 17(4), 171–178. <https://doi.org/10.3923/pjn.2018.171.178>
- Ehlers, J. D., & Hall, A. E. (1998). Heat tolerance of contrasting cowpea lines in short and

- long days. *Field Crops Research*, 55(1–2), 11–21. [https://doi.org/10.1016/S0378-4290\(97\)00055-5](https://doi.org/10.1016/S0378-4290(97)00055-5)
- Ekesa, B., Walingo, M., & Onyango, M. (2009). Accesibility to and consumption of indigenous vegetables and fruits by rural households in Matungu division, western Kenya. *African Journal of Food, Agriculture, Nutrition and Development*, 9(8). <https://doi.org/10.4314/ajfand.v9i8.48410>
- Gomez, C. (2004). *COWPEA Post-harvest Operations. Food and Agriculture Organisation Of The United Nations (FAO), Rome, Italy*. 6–8.
- Gonçalves, A., Goufo, P., Barros, A., Domínguez-Perles, R., Trindade, H., Rosa, E. A. S., ... Rodrigues, M. (2016). Cowpea (*Vigna unguiculata* L. Walp), a renewed multipurpose crop for a more sustainable agri-food system: Nutritional advantages and constraints. *Journal of the Science of Food and Agriculture*, 96(9), 2941–2951. <https://doi.org/10.1002/jsfa.7644>
- Hewett, E. W. (2006). An overview of preharvest factors influencing postharvest quality of horticultural products. *International Journal of Postharvest Technology and Innovation*, 1(1), 4–15. <https://doi.org/10.1504/IJPTI.2006.009178>
- Hornick, S. B. (1992). Factors affecting the nutritional quality of crops. *American Journal of Alternative Agriculture*, 7(1–2), 63–68. <https://doi.org/10.1017/S0889189300004471>
- Khalid, I. I., & Elhardallou, S. B. (2016). Factors That Compromise the Nutritional Value of Cowpea Fluor and Its Protein Isolates. *Food and Nutrition Sciences*, 07(02), 112–121. <https://doi.org/10.4236/fns.2016.72013>
- Lee, S. K., & Kader, A. A. (2000). Preharvest and postharvest factors influencing vitamin C content of horticultural crops. *Postharvest Biology and Technology*, 20(3), 207–220. [https://doi.org/10.1016/S0925-5214\(00\)00133-2](https://doi.org/10.1016/S0925-5214(00)00133-2)
- Modi, A. T. (2007). Growth temperature and plant age influence on nutritional quality of Amaranthus leaves and seed germination capacity. *Water SA*, 33(3 SPECIAL EDICTION), 369–375. <https://doi.org/10.4314/wsa.v33i3.180598>
- Muhanji, G., Roothaert, R. L., Webo, C., & Stanley, M. (2011). African indigenous vegetable enterprises and market access for small-scale farmers in East Africa. *International Journal of Agricultural Sustainability*, 9(1), 194–202. <https://doi.org/10.3763/ijas.2010.0561>
- Nangula P. Uusiku, A. O. Andre´ Oelofse, Kwaku G. Duodu, Megan J. Bester, Mieke Faber (2010). Nutritional value of leafy vegetables of sub-Saharan Afric and their potential contribution to human health: A review. *Journal of Food Composition and Analysis*, 499-509.
- Nyadanu, D., & Lowor, S. T. (2014). Promoting competitiveness of neglected and underutilized crop species: comparative analysis of nutritional composition of indigenous and exotic leafy and fruit vegetables in Ghana. *Genetic Resources and Crop Evolution*, 62(1), 131–140. <https://doi.org/10.1007/s10722-014-0162-x>
- OECD. (2016). *Safety Assessment of Transgenic Organisms in the Environment, Volume 6: Consensus Documents, Harmonisation of Regulatory oversight in biotechnology*. <https://doi.org/10.1787/9789264253018-en>
- Ohler, T. A., Nielsen, S. S., & Mitchell, C. A. (1996). Varying plant density and harvest time to optimize cowpea leaf yield and nutrient content. *HortScience*, 31(2), 193–197. <https://doi.org/10.21273/hortsci.31.2.193>
- Okello, J., Hutchinson, M., Mwang'ombe, A., Ambuko, J., Olubayo, F., & Mwakangalu, M. (2015). Consumer Demand for Value-added Products of African Indigenous Vegetables in Coastal Kenya: The Case of Sun-dried and Frozen Cowpea Leaves. *Journal of Agriculture, Food Systems, and Community Development*, 189–207. <https://doi.org/10.5304/jafscd.2015.061.004>
- Onyango, C. M., Imungi, J. K., & Harbinson, J. (2008). The Physico-Chemical Characteristics and Some Nutritional Values of Vegetable Amaranth Sold in Nairobi-Kenya The Physico-Chemical Characteristics and Some Nutritional Values of. *Ecology*

- of *Food and Nutrition*, (November 2018). <https://doi.org/10.1080/03670240802003926>
- Paul van Jaarsvela, M. F. Mieke Faber, Ina van Heerden, Friede Wenhold, Willem Jansen van Rensburg d, Wim van Averbek. (2014). Nutrient content of eight African leafy vegetables and their potential contribution to dietary reference intakes. *Journal of Food Composition and Analysis*, 77-84.
- Pietruszka, M., Lewicka, S., & Pazurkiewicz-Kocot, K. (2007). Temperature and the growth of plant cells. *Journal of Plant Growth Regulation*, 26(1), 15–25. <https://doi.org/10.1007/s00344-006-0023-8>
- Rickman, J. C., Bruhn, C. M., & Barrett, D. M. (2007). Nutritional comparison of fresh , frozen , and canned fruits and vegetables II . Vitamin A and carotenoids , vitamin E , minerals and fiber. *Journal of the Science Of Food and Agriculture*, 1196(October 2006), 1185–1196. <https://doi.org/10.1002/jsfa>
- Rodríguez, V. M., Soengas, P., Alonso-Villaverde, V., Sotelo, T., Cartea, M. E., & Velasco, P. (2015). Effect of temperature stress on the early vegetative development of Brassica oleracea L. *BMC Plant Biology*, 15(1), 1–9. <https://doi.org/10.1186/s12870-015-0535-0>
- Sebetha, E. T., Ayodele, V. I., Kutu, F. R., & Mariga, I. K. (2010). Yields and protein content of two cowpea varieties grown under different production practices in Limpopo province , South Africa. *African Journal of Biotechnology*, 9(5), 628–634.
- Sebetha, E. T., Modi, A. T., & Owoeye, L. G. (2014). Cowpea Crude Protein as Affected by Cropping System, Site and Nitrogen Fertilization. *Journal of Agricultural Science*, 7(1). <https://doi.org/10.5539/jas.v7n1p224>
- Sinha, R., & Kawatra, A. (2003). Effect of processing on phytic acid and polyphenol contents of cowpeas [*Vigna unguiculata* (L) Walp]. *Plant Foods for Human Nutrition*, 58(3), 1–8. <https://doi.org/10.1023/B:QUAL.0000040322.01063.d4>
- Steyn, N. P., Olivier, J., Winter, P., Burger, S., & Nesamvuni, C. (2001). A survey of wild, green, leafy vegetables and their potential in combating micronutrient deficiencies in rural populations. *South African Journal of Science*, (August), 276–278.
- Timko, M. P., & Singh, B. B. (2008). Cowpea , a Multifunctional Legume Cowpea , a Multifunctional Legume. In P. H. Moore & R. Ming (Eds.), *Genomics of Tropical Crop Plants* (pp. 227–258). <https://doi.org/10.1007/978-0-387-71219-2>
- Verkerke, W., Labrie, C., & Dueck, T. (2015). The effect of light intensity and duration on Vitamin C concentration in tomato fruits. *Acta Horticulturae*, 1106(Favhealth 2014), 49–53. <https://doi.org/10.17660/ActaHortic.2015.1106.8>
- Wojciech J. Florkowski, R. L. Robert L. Shewfelt, Bernhard Brueckner and Stanley E. Prussia. (2009). *Postharvest Handling: A Systems Approach*. Elsevier Inc. Academic Press.
- Xiong, H., Shi, A., Mou, B., Qin, J., Motes, D., Lu, W., ... Wu, D. (2016). Genetic diversity and population structure of cowpea (*vigna unguiculata* l. walp). *PLoS ONE*, 11(8), 1–15. <https://doi.org/10.1371/journal.pone.0160941>