

## Evaluation of the Effect of Planting Distance and Harvesting Time on the Carotenoids and Phytochemicals of Selected Orange-Fleshed Sweet Potato Varieties

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**Keywords:** Orange Fleshed sweet potatoes Umuspo 3, OFSP Ex-Igbariam, Planting space, Harvesting period, Carotenoid, Phytochemicals.

**Abstract.** We evaluated the carotenoid profile and concentration (by HPLC) and the phytochemical content of two OFSP varieties (Umuspo 3 and Ex-Igbariam) planted at three distances (20 cm, 30 cm and 40 cm) and harvested in two different periods (12<sup>th</sup> and 16<sup>th</sup> weeks after planting) respectively. Carotene contents of the outer peel and inner flesh of the sweet potato varieties were also determined. The results showed wide variation in the carotenoid and phytochemical content among the varieties at different planting spaces and harvest periods. Umuspo 3 planted at 20 cm, 30 cm and 40 cm had significantly greater carotenoid concentration than Ex-Igbariam variety. The predominant carotenoid was  $\beta$ -carotene with highest concentration obtained from 40 cm planting distance (92.82 $\mu$ g/g) and 30 cm (80.97 $\mu$ g/g) for Umuspo 3. Ex-Igbariam at 30 cm planting distance contained 2.51 $\mu$ g/g  $\beta$ -carotene when harvested after 16<sup>th</sup> weeks. Also the highest  $\beta$ -carotene concentration was from Umuspo 3 flesh sample planted 30 and 40 cm (409.45 and 441.15 mg/100g) and the peel for samples planted 30 and 40 cm (490.47 and 640.69 mg/100g, respectively) at the 12<sup>th</sup> week of harvest. Flavonoids were present in significant amounts (310.62mg/100g) in Umuspo 3 planted at 30 cm and harvested after 12<sup>th</sup> week while in total polyphenol, significant quantities of  $\approx$ 42.12mg/100g was present in Ex-Igbariam spaced at 30 cm and 40 cm and harvested after 16<sup>th</sup> week. Provitamin A carotenoid was calculated and Umuspo 3 pro-vitamin A carotenoid was significantly higher ( $p < 0.05$ ) with highest concentration (742.26 RE/100g) present in samples from 40 cm planting distance. The results showed that planting space and harvesting period had significant impact on the carotenoid and phytochemical concentrations of OFSP varieties. Planting distances of 30 and 40 cm are recommended for high carotenoid content in the two sweet potato varieties.

### Introduction

Sweet potato is an important root crop in the tropical and subtropical regions of the world [1, 2]. It is a major food staple which contributes substantially to national food security. As a result, it is used by rural and urban population in fresh and processed forms to supply energy and some micronutrients, particularly the pro-vitamin A carotenoid [3]. Sweet potatoes require fewer inputs and can tolerate marginal growing conditions such as dry or poor soil and yet produce good yields with less labour.

Nigeria is one of the major producers of sweet potato in West Africa [4]. Despite this, sweet potato has received limited research attention especially as a source of carotenoid and other micronutrient, and is generally underutilized. Sweet potato as an untapped nutrient-dense staple crop has the potential to meet the dietary vitamin A requirement and can increase the absorption of zinc and iron to meet the physiological need of the vulnerable group of the Nation's population to reduce the burden of micro-nutrient deficiency. In African, it is estimated that 43million children under the age of five are threatened by vitamin A deficiency (VAD), a condition that causes blindness and premature death [5].

Orange-fleshed sweet potato is high in beta-carotene (a pre-cursor of vitamin A) and can be used as a food based approach in combating vitamin A deficiency. This variety of sweet potato has the advantage of substituting the expensive vitamin A rich animal foods to meet the daily requirement of vitamin A. Vitamin A deficiency remains a serious public health problem in developing countries. Beside the supply of  $\beta$ -carotene, orange-fleshed sweet potato is also rich in polyphenolic compounds, minerals and vitamins which group them as functional food for the improvement of human health [6]. Polyphenolic compounds from sweet potato have several physiological benefits like protection against stomach cancer, leukemia [7], improves diabetic condition in humans [8] and as bio-fortified crop, can combat malnutrition in small and marginal farming communities. The antioxidant activity of sweet potato has been attributed to the constituent phenolic compounds [9].

Phenolic compounds function as scavengers of free-radical molecules inside the cell. In humans, the damage caused by free radicals has been implicated in the development of cancer, cataracts and heart disease [10]. With high micro-nutrient content and phenolic compounds (chlorogenic acid, caffeic acid and three isomers of the dicaffeoylquinic acids), significant contributions to the nutraceutical function of sweet potato can be canvassed [10].

Optimal yield and low cost are two most important drivers of food production all over the world. Etela and Kalio [11] studied the optimal yield components of three sweet potato varieties and observed that root yields were higher at 20 weeks after harvest than at 12 weeks after harvest. Agricultural practices and tuber yield, environmental factors and genotypes are always considered by researchers but not their influence on the biochemical composition and superior nutritive value to improve human health [12]. Improved nutrition in OFSP lines with regards to  $\beta$ -carotene can be achieved if high yielding  $\beta$ -carotene OFSP is selected with important agronomic practice, such as planting space and harvesting timing. Therefore, assessment of biochemical composition of orange fleshed sweet potato genotypes is essential for selecting the cultivar having high amounts of  $\beta$ -carotene and other functional phytonutrients beneficial to the physiological need of humans. The relevance of orange-fleshed sweet potatoes in alleviating vitamins A (Pro-vitamin A) deficiency is complementary to this study and therefore, assessment of farming practices involving planting space and harvesting periods of the tubers of different cultivars of orange-fleshed sweet potatoes were made in order to select proper farming conditions that will maximize nutrient yield.

The major objective of this work was to assess ideal planting space and maturity period that will lead to optimal phyto-nutrients and carotenoid concentration in orange-fleshed sweet potatoes thereby determining optimal conditions for planting and harvesting developed OFSP varieties for nutrient content maximization. In this study, vines of two cultivars of orange-fleshed sweet potato were cultivated with different planting spaces, the tubers were harvested at two different dates and the phyto-nutrient and carotenoid composition of their tubers were evaluated

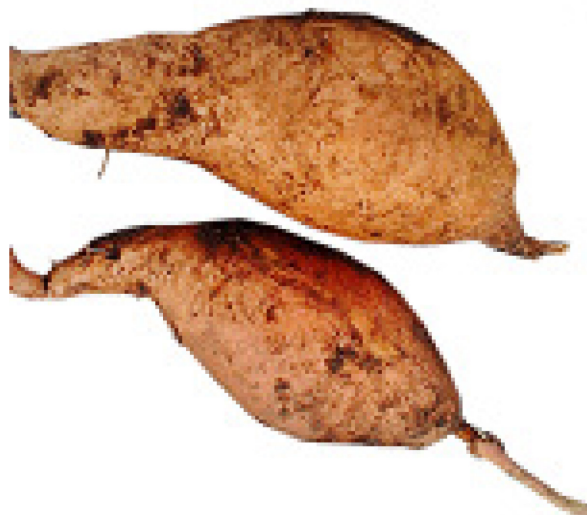
## Materials and Methods

*Plant materials:* Two orange-fleshed sweet potato varieties (Ex-Igbariam and Umuspo 3) planted 30 cm x 1m and 40 cm x 1m and harvested at 12<sup>th</sup> and 16<sup>th</sup> weeks as shown in Fig. 1a and 1b were used for the research analysis.

Sweet potato varieties (Umuspo 3 and Ex-Igbariam) were planted at the National Root Crops Research Institute experimental farm, Umudike, Abia State, Nigeria. Umudike which is situated between latitude 05°29N and longitude 07°33 E and 122m altitude has its soil classified as sandy loam, acidic and characterized as an ultisol. The two different varieties were planted using three different planting spaces: 20cm x 1m, 30cm x 1m and 40cm x 1m on rows and between lines respectively. The treatments were arranged as split plot in randomized complete block design with three replications. The blocks of experimental units were uniform so that the observed differences between treatments will be largely due to true differences between treatments. Samples from each variety were harvested at 12<sup>th</sup> and 16<sup>th</sup> weeks after planting and analyzed.



**Fig. 1a.** Ex-Igbariam Sample.



**Fig. 1b.** Umuspo 3 Sample.

### *Field Work, Experimental Design and Treatments*

#### *Determination of Flavonoid content*

The flavonoid content was determined by the gravimetric method as described by Harbone and Williams [13]. Five grams of the powdered sample was placed into a conical flask and 50 mL of water and 2 mL HCl solution was added. The solution was allowed to boil for 30 min. The boiled mixture was allowed to cool before it was filtered through Whatman filter paper (No.42). Ten (10) mL of ethyl acetate extract which contained flavonoid was recovered while the aqueous layer was discarded. A pre-weighed Whatman filter paper (No. 42) was used to filter the second ethyl-acetate layer, the residue was then placed in an oven to dry at 60°C. It was cooled in a desiccator, weighed and the quantity of flavonoid was determined.

#### *Determination of total polyphenol content*

Three (3) gram of the sample was put into a 100mL conical flask and macerated with 50 mL of acidified methanol and filtered. Twenty five (25) mL of the filtrate was pipette and added to 0.125 mL Folin-Ciocalteu reagent with 0.1mL 0.5M aqueous Sodium hydroxide and was allowed to stand in the dark at room temperature for 15mins. The absorbance was measured at 650nm in a spectrophotometer (BIOBASE LCD BK-UV1800PC). The concentration of total polyphenol was calculated.

#### *Determination of Carotenoids content*

Carotenoids analysis of the sweet potato samples was carried out at the Crop Utilization Unit (Laboratory) of International Institute of Tropical Agriculture (IITA) Ibadan.

#### *Sample Preparation*

The sweet potatoes were selected randomly and washed with clean water to remove adhering soil particles; it was peeled, washed and quartered longitudinally. Two opposite sections were taken and sliced into small pieces (1 cm) and mixed manually. They were packaged in aluminum foil, labeled and stored at -18°C in a deep freezer.

#### *Carotenoid extraction*

Carotenoid was extracted according the method of Rodriguez-Amaya and Kimura [14]. About 3 g of each sample was ground in a laboratory ceramic mortar and pestle with about 50 mL of cold acetone. The ground sample was filtered in a Buchner funnel equipped with a filter paper (Whatman No. 42 filter paper). The residue was returned to the mortar and the extraction was repeated using 20 mL acetone until the residue was nearly colourless. The total extract was transferred to a separating funnel (250 mL) containing 20 mL of petroleum ether (boiling point = 40°C to 60°C).

One liter of distilled water was used to wash the organic phase which separated from the aqueous phase. The aqueous phase was discarded. The organic phase was again washed with dilute brine solution to break-up any emulsions that may have formed. The brine solution which separated from the organic phase was discarded. The organic phase was collected through anhydrous Sodium sulphate (15 g) into a 25 mL flat bottom flask. Ten (10) mL of the sample extract was concentrated with a rotary evaporator (Buchi Waterbath B-481 Switzerland) and dried under vacuum for reverse-phase HPLC determination of the various carotenoids.

The remaining 15 mL sample extract was used to determine the total carotenoid at 450 nm in a spectrophotometer (BIOBASE LCD BK-UV1800P). The values of carotenoid obtained were multiplied by 100 to give the carotenoid content in  $\mu\text{g}/100\text{ g}$ . Sample preparation, extraction and analysis were performed under protected white fluorescent lighting.

#### *Carotenoid quantification*

HPLC procedure for carotenoid analysis in the OFSP sweet potato varieties was adopted from Howe and Tanumihardjo [15]. The carotenoid sample was reconstituted in methanol/dichloroethane mixture (1000 $\mu\text{l}$ , 50:50v/v). Ten (10)  $\mu\text{l}$  was injected into the HPLC consisting of a guard column, C30 YMC carotenoid column (4.6x250 mm, 3 $\mu\text{m}$ ), 625 HPLC pump, 717 auto sampler and a 2996 photodiode array detector (Waters Corporation, Milford, M.A). The isocratic elution was 50% solvent A (100% methanol) and 50% solvent B (100% methyl tert-butyl ether) at 1mL/min. for 15 minutes.  $\beta$ -carotene, the major carotenoid eluted at about 6 minutes. Absorbance of eluents from chromatograms were read automatically at 450 nm and the identification of different carotenoid was done using standards, and with the verification of the absorption spectrum. The quantification of different carotenoid was done by the integration of the peak areas against respective standard curves.

#### *Determination of $\beta$ -carotene content of peel and flesh portions of OFSP varieties*

The  $\beta$ -carotene content of the peel and flesh portions of the OFSP varieties were determined according to AOAC [16]. Forty (40) mL sample was taken into 1000 mL beaker and 20 mL of acetone was added. The addition of acetone continued until the mixture was saturated and the weight recorded. The sample was covered and stored in the refrigerator overnight. The aqueous layers were extracted by using pipette and discarded while the remaining content was weighed and recorded. Water contained in the sample was filtered using a filter funnel with filter paper. The remaining solid substance (mashed potato) was placed in another beaker and 20 mL acetone was added. About 15g mixed sample was weighed and placed in a filtration funnel. Then 2 mL of acetone and 15mL of  $\text{CH}_2\text{Cl}_2$  was added to facilitate solubility and filterability, the mixture was filtered using vacuum filtration method. The  $\text{CH}_2\text{Cl}_2$  was removed and 3 drops of  $\text{CaCl}_2$  was added to enhance the complexation of organic compounds. The samples were put into pre-weighed and moderately heated vials. One (1mL) Petroleum ether was added and further purified by column chromatography. Air was pumped into the content to seal off hexane, sand and petroleum ether.  $\beta$ -carotene and hexane were captured in a test tube. The absorbance at 450nm was read in quartz cells UV-VIS spectrophotometer. Reading was taken quickly, since petroleum ether is a volatile solvent. The  $\beta$ -carotene concentration (C in mg.l-1) was calculated according to Beer –Lamberts law from measured data of the absorbance.

## **Results and Discussion**

### *Carotenoid concentration and profile of OFSP (Ex-Igbariam and Umuspo 3) varieties*

#### *Data Analysis*

Data were analyzed with Statistical Analysis Software program (SAS 9.0) using one way analysis of variance (ANOVA). Least significant differences were calculated at  $p < 0.05$ . Mean comparison was made for all the significant treatments and mean  $\pm$  standard deviations are presented in the tables of results.

**Table 1.** Carotenoid profile and concentration of orange-fleshed sweet potato (*Ipomea batatas*) varieties (Umuspo 3 and Ex-Igbariam) at different planting distances.

Samples	Xanthophylls			Cyclic carotenes					
	Lutein	Zeaxanthin (µg/g)	B-cryptoxanthin (µg/g)	α-Carotene (µg/g)	13-Cis-β-Carotene (µg/g)	Trans-β-carotene (µg/g)	9-Cis-β-carotene (µg/g)	Total β-carotene (µg/g)	vitamin A (RE/100g)
V4S1	0.29 <sup>f</sup>	0.27 <sup>f</sup>	0.11 <sup>f</sup>	0.23 <sup>f</sup>	0.9 <sup>f</sup>	0.26 <sup>e</sup>	0.08 <sup>f</sup>	0.44 <sup>f</sup>	17.40
V4S2	0.85 <sup>e</sup>	1.69 <sup>c</sup>	0.42 <sup>d</sup>	0.58 <sup>e</sup>	0.28 <sup>d</sup>	1.88 <sup>d</sup>	0.35 <sup>d</sup>	2.51 <sup>d</sup>	30.84
V4S3	0.95 <sup>d</sup>	1.27 <sup>e</sup>	0.29 <sup>e</sup>	0.60 <sup>d</sup>	0.15 <sup>e</sup>	0.47 <sup>e</sup>	0.25 <sup>e</sup>	0.88 <sup>e</sup>	18.30
V1S1	1.72 <sup>c</sup>	1.36 <sup>d</sup>	4.35 <sup>c</sup>	2.99 <sup>e</sup>	4.57 <sup>b</sup>	62.09 <sup>c</sup>	1.77 <sup>b</sup>	68.43 <sup>c</sup>	591.42
V1S2	2.23 <sup>b</sup>	2.36 <sup>b</sup>	4.88 <sup>b</sup>	3.29 <sup>b</sup>	2.08 <sup>c</sup>	77.91 <sup>b</sup>	0.97 <sup>c</sup>	80.97 <sup>b</sup>	602.10
V1S3	2.79 <sup>a</sup>	3.23 <sup>a</sup>	7.83 <sup>a</sup>	3.62 <sup>a</sup>	6.02 <sup>a</sup>	84.81 <sup>a</sup>	1.98 <sup>a</sup>	92.82 <sup>a</sup>	742.26
LSD <sub>(0.05)</sub>	0.0167	0.0241	0.0749	0.0659	0.1135	0.3119	0.0374	0.3046	

Means with different superscript (abcdef) within the same column are significantly different ( $p < 0.05$ ) Values are means  $\pm$  standard deviation of three replicate

Sample code: V4S1–Ex-igbariam 20cm, V4S2–Ex-igbariam 30cm, V4S3–Ex-igbariam 40cm, V1S1–Umuspo 3 20cm, V1S2–Umuspo 3 30cm, V1S3–Umuspo 3 40cm

The effect of planting space and harvesting period on the carotenoid concentration and profile of OFSP (Umuspo 3 and Ex-Igbariam varieties) are shown in Table 1 and Fig. 2 (chromatograms) respectively. The results showed that the carotenoid concentration varied between the two varieties. It was evident that a large concentration of the identified carotenoid was  $\beta$ -carotene comprising of All-trans  $\beta$ -carotene, 13-cis  $\beta$ -carotene, 9-cis  $\beta$ -carotene with Umuspo 3 variety showing a far richer source of carotenoid than Ex-Igbariam variety. In general, the mean lutein content was  $1.47 \mu\text{g/g}$  and varied between  $0.29 \mu\text{g/g}$  and  $2.77 \mu\text{g/g}$ . Umuspo3 (V1S1, V1S2 and V1S3) samples had higher values of lutein than Ex-Igbariam (V4S1, S4S2 and S4S3) samples irrespective of spacing distance. It was observed that samples of both varieties planted at 40cm apart in rows had higher lutein content than those of other planting spaces.

Umuspo3 (V1S3) had the highest zeaxanthin concentration ( $3.23 \mu\text{g/g}$ ) which was significantly different from all other samples. Ex-Igbariam (V4S1) had the least value ( $0.27 \mu\text{g/g}$ ). Umuspo 3 variety (V1S1, V1S2 and V1S3) maintained higher zeaxanthin values than Ex-Igbariam lines (V4S1, S4V2 and S4V3) respectively.

The cryptoxanthin value of Umuspo 3 (V1S3) ( $7.83 \mu\text{g/g}$ ) was higher than the other samples. It was observed that Ex-Igbariam samples had less cryptoxanthin values than Umuspo 3 values.

The same trend was observed in  $\alpha$ -carotene. Umuspo 3 V1S3 ( $3.62 \mu\text{g/g}$ ), V1S2 ( $3.29 \mu\text{g/g}$ ) and V1S1 ( $2.99 \mu\text{g/g}$ ) maintained higher values than Ex-Igbariam samples. It was observed that Umuspo-3 is a rich source of  $\alpha$ -carotene which can contribute about 50 % to pro-vitamin content.

Umuspo 3 V1S3 planted at 40 cm contained highest quantity of 13-cis- $\beta$ -carotene ( $6.02 \mu\text{g/g}$ ) while the lowest concentration ( $0.15 \mu\text{g/g}$ ) was observed in Ex-Igbariam V4S3. The variation in Umuspo3 13-cis- $\beta$ -carotene over that of Ex-Igbariam was highly significant.

All trans- $\beta$ -carotene was rich in Umuspo 3 V1S3 ( $84.81 \mu\text{g/g}$ ). Samples Ex-Igbariam V4S1 and V4S3 had the least values of All trans- $\beta$ -carotene. Trace amount of 9-cis- $\beta$ -carotene was detected in Ex-Igbariam V4S1 while Umuspo 3 V1S3 had the highest value ( $1.98 \mu\text{g/g}$ ).

Ex-Igbariam spacing lines generally had the least values for the total  $\beta$ -carotene. Umuspo V1S3 had the highest total  $\beta$ -carotene concentration ( $92.82 \mu\text{g/g}$ ) which was significantly different from other samples while Ex-Igbariam V4S3 had the least concentration ( $0.44 \mu\text{g/g}$ ).

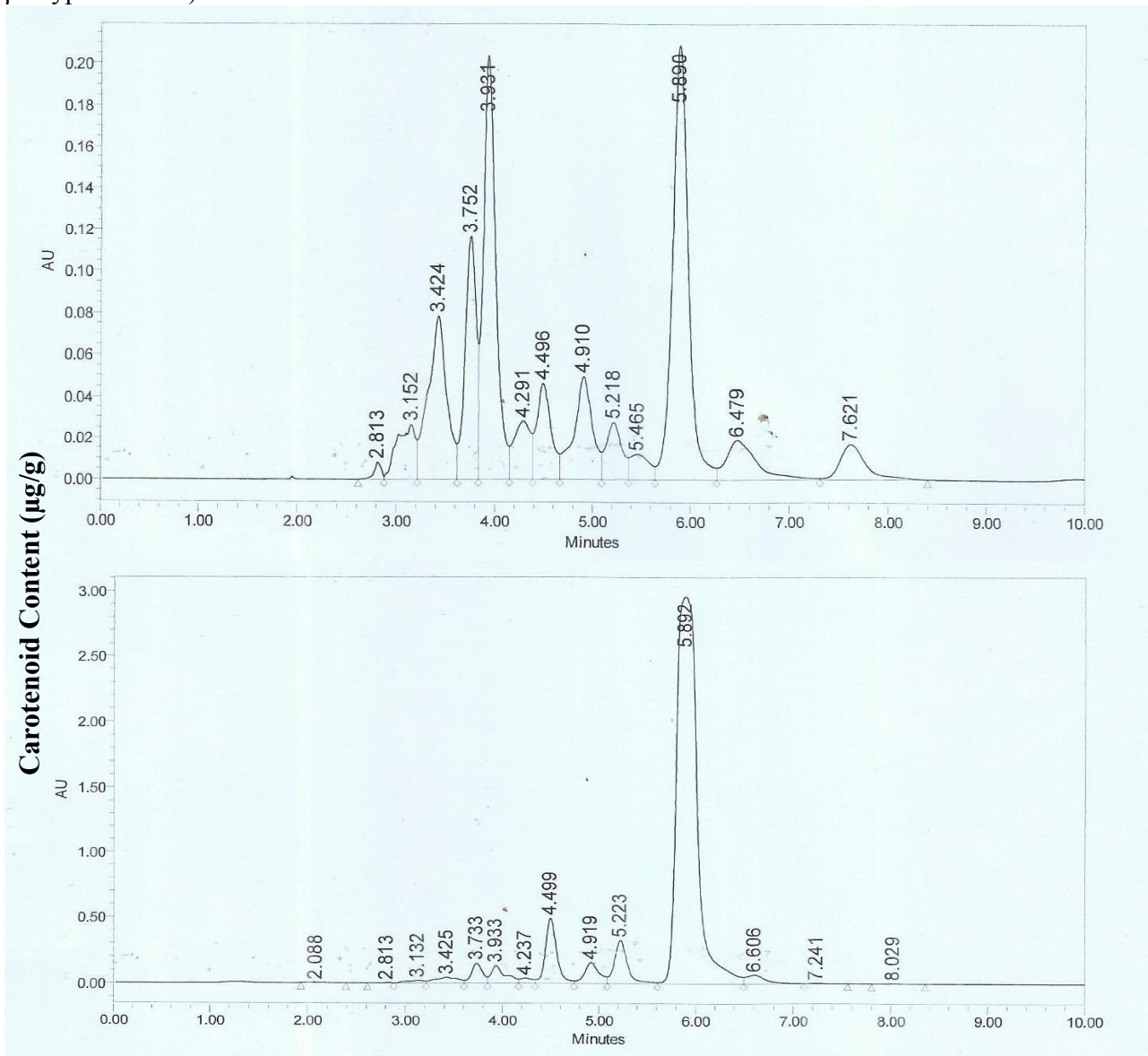
The concentration of carotenoid obtained in this study showed that Umuspo 3 variety irrespective of vine spacing was higher than those of Ex-Igbariam. The highest value was observed in Umuspo 3 V1S3 (40 cm spacing), while in Ex-Igbariam, the highest value was observed in V4S2 (30 cm spacing). Widening the planting spaces as observed in 30cm x 1m and 40 x 1m increased

the carotenoids content of the tubers. This may be attributed to lesser biomass canopy when compared to 20 cm x 1m resulting in higher radiation interception at 450-550 nm and increased photosynthetic activity, and thus, may have directly affected carotenogenesis. In another report, Loebenstein and Thottappilly [17] had suggested 1m x 1m planting space for sweet potato varieties based on their agro-ecological environment and farming practice, however, the planting spaces in this study are suitable for Umudike-Nigerian ultisol, classified as acidic and sandy loam.

Comparing with agricultural practice as influenced by date of harvesting, Etela and Kalio [11] reported that root yields were higher at 20 weeks after harvest than at 12 weeks. On carotenoid yield of sweet potato varieties, Ukom et al. [3, 18] reported lower total  $\beta$ -carotene concentration ( $\leq 18.10\mu\text{g/g}$ ) and ( $\leq 69.42\mu\text{g/g}$ ) than the values for Umuspo 3 lines ( $\leq 92.82\mu\text{g/g}$ ). Findings reported in different types of root and tuber crops [19, 20-22] show that their carotenoids concentration are considerably lower than the values of Umuspo 3 variety. Although analytical results are for a particular staple grown in a particular geo-ecological area, such varying carotenoid concentrations results depicts the environmental, cultural and genetic factors.

Table 1 also shows the vitamin A (Retinol equivalent 1RE/100g) potentials of the sweet potato varieties. All trans- $\beta$ -carotene, the major provitamin A carotenoid, its isomers,  $\alpha$ -carotene and  $\beta$ -cryptoxanthin were used to calculate the Retinol Equivalent according to FAO/WHO [23].

One Retinol equivalent (1RE) =  $6 \times \text{trans-}\beta\text{-carotene} + 12(\text{cis-}\beta\text{-carotene} + \alpha\text{-carotene} + \beta\text{-cryptoxanthin})$ .



Figures 2a and 2b. Chromatograms of Ex-Igbariam (30cm) and Umuspo 3 (40cm).

The result showed that Umuspo3 planting spaces contributed significantly to vitamin A (retinol) content, ranging from 591.42 to 742.26 RE/100g when compared to Ex-Igbariam, which ranged from 17.40 to 30.84 RE/100g. This implies the ability of Umuspo 3 variety in contributing to reducing vitamin A deficiency that is endemic in sub-Saharan African. The provitamin A value of Umuspo 3 samples can be compared with the report of Paul et al. [24] for different varieties of orange-fleshed sweet potato (values ranging from 694RE/100g to 948 RE/100g). According to K'osambo et al. [25] the provitamin A value of CIP440078, a white variety of sweet potato was 6.6 RE/100g and Zapallo (CIP420027) pale orange was 258.2RE/100g. These values are lower than the values reported in this study for both Umuspo 3 and Ex-Igbariam samples. Since the daily recommended intake of vitamin A is 900 $\mu$ g/day and 400 $\mu$ g/day for adults and children below 5years respectively, it means that the pro-vitamin A values generated from the Umuspo-3 lines can meet the daily recommended intake among the vulnerable groups of the population.

Figs. 2a and 2b are the chromatograms of Ex-Igbariam planted at 30 cm (S4V2) and Umuspo3 spaced at 40 cm (V1S3) apart in rows. They are the graphical representation of the absorption peak heights against retention time at 450 nm wavelength. Fig. 2a shows the chromatogram of Ex-Igbariam variety planted at 30cm (S4S2). Thirteen carotenoid fractions eluted at different retention times were observed. Seven major peaks were identified while six minor peaks believed to be xanthophylls were not identified due to lack of internal standards. Similar findings have been reported in different types of crops [20, 22, 26]. The different identified carotenoid peaks are labeled in their respective chromatograms. Fig. 2b show the chromatogram of Umuspo3 variety planted at 40 cm (V1S3) apart in rows. Fourteen carotenoid fractions eluted at different retention times were observed. Seven major peaks were also identified. Trans- $\beta$ -carotene (peak 6) predominated with 84.81 $\mu$ g/g indicating 75.76% carotenoid concentration, followed by  $\beta$ -cryptoxanthin (peak 3) 7.83 $\mu$ g/g (6.69%), 13-Cis- $\beta$ -carotene (peak 5) 6.02 $\mu$ g/g (5.36%),  $\alpha$ -carotene (peak4) 3.62 $\mu$ g/g (2.93%), zeaxanthin (peak 2) 3.23 $\mu$ g/g (2.43%), lutein (peak 1) 2.79 $\mu$ g/g (2.12%), and 9-Cis- $\beta$ -carotene (peak 7)1.98 $\mu$ g/g (1.69%) respectively.

#### *$\beta$ -Carotene composition of the flesh and peel of the OFSP varieties*

Table 2 shows the  $\beta$ -carotene composition of the flesh and peel of Umuspo 3 and Ex-Igbariam varieties planted 20cm, 30cm and 40 cm and harvested at 12<sup>th</sup> and 16<sup>th</sup> weeks maturity.

**Table 2.**  $\beta$ -carotene Composition of the flesh and peel of OFSP varieties.

Samples	Flesh (mg/100g)		Peel (mg/100g)	
	12weeks	16weeks	12weeks	16weeks
V4S1	135.83 <sup>d</sup> ±2.56	163.65 <sup>d</sup> ±3.09	267.56 <sup>e</sup> ±1.19	295.12 <sup>e</sup> ±2.94
V4S2	133.17 <sup>d</sup> ±2.42	160.45 <sup>d</sup> ±2.92	285.54 <sup>d</sup> ±3.65	316.09 <sup>d</sup> ±2.99
V4S3	132.90 <sup>d</sup> ±6.62	161.41 <sup>d</sup> ±7.55	277.28 <sup>de</sup> ±5.44	308.09 <sup>d</sup> ±6.04
V1S1	409.45 <sup>b</sup> ±6.27	329.86 <sup>b</sup> ±2.42	441.15 <sup>c</sup> ±1.20	475.42 <sup>b</sup> ±2.81
V1S2	490.47 <sup>a</sup> ±4.16	396.89 <sup>a</sup> ±4.11	640.67 <sup>a</sup> ±2.49	487.27 <sup>a</sup> ±6.90
V1S3	378.54 <sup>c</sup> ±3.15	225.94 <sup>c</sup> ±1.82	475.26 <sup>b</sup> ±2.65	359.01 <sup>c</sup> ±6.27

Means with different superscript (abcd) within the same column are significantly different ( $p < 0.05$ ) Values are means  $\pm$  standard deviation of three replicate

Sample code: V4S1–Ex-igbariam 20cm, V4S2–Ex-igbariam 30cm, V4S3–Ex-igbariam 40cm, V1S1–Umuspo 3 20cm, V1S2–Umuspo 3 30cm, V1S3–Umuspo 3 40cm.

Table 2 shows the  $\beta$ -carotene content of the peel and flesh of OFSP varieties planted at 20cm, 30cm and 40cm and harvested at 12<sup>th</sup> and 16<sup>th</sup> weeks. Significant variations ( $p < 0.05$ ) existed among some of the samples, especially for Umuspo 3 harvested at 12<sup>th</sup> and 16<sup>th</sup> weeks. Umuspo 3 V1S2 irrespective of harvesting period (that is after 12<sup>th</sup> and 16<sup>th</sup> weeks) exhibited the highest  $\beta$ -carotene values for both flesh and peel. At 16<sup>th</sup> week, Umuspo 3 V1S2 flesh had the highest value (396.89mg/100g) while Ex-Igbariam V4S2 had the least value (160.45mg/100g). Approximately, 19% increase was observed in Ex-Igbariam spacing lines at 16<sup>th</sup> week while Umuspo3 flesh showed a decrease in  $\beta$ -carotene at 16<sup>th</sup> week harvest.

At 12<sup>th</sup> of week harvest, Umuspo 3 V1S2 peel had the highest value (640.67mg/100g) of  $\beta$ -carotene while Ex-Igbariam V4S1 had the least value (267.56mg/100g). At 16<sup>th</sup> week, Umuspo 3 V1S2 had the highest value (487.27mg/100g) which was significantly different from other samples while sample V4S1 had the least value (295.12mg/100g).

Whereas the  $\beta$ -carotene content of Ex-Igbariam samples increased at 16<sup>th</sup> week of harvest, it was not so for Umuspo 3 samples which showed decreases at 16<sup>th</sup> week with the exception of V1S1 which gained 7.5% increase. It was observed that Umuspo 3 OFSP variety had higher  $\beta$ -carotene content than Ex-Igbariam variety and varied significantly amongst themselves at both 12<sup>th</sup> and 16<sup>th</sup> weeks. Planting space affected the  $\beta$ -carotene content of samples as there was variation at the different planting space for both varieties. At 30cm planting space, the  $\beta$ -carotene content of both varieties was higher than those of other planting spaces as seen in samples V4S2 and V1S2, demonstrating the effect of increased carotenoids biosynthesis due possibly to higher photosynthetic activity and increased radiation interception. In all considerations, both the flesh and peel of these OFSP varieties are rich sources of carotenoids to benefit retinol bioavailability and human nutrition.

#### *Phytochemical content of OFSP (Ex-Igbariam and Umuspo 3) varieties*

Table 3 shows the influence of harvesting time on the phytochemical content of sweet potato varieties. Significant differences ( $p < 0.05$ ) existed among planting spaces and harvesting periods in all parameters tested.

**Table 3.** Effect of harvesting time on the phytochemical composition of sweet potato (*Ipomea batatas*) varieties.

Samples	Flavonoids(mg/100g)		Total polyphenol(mg/100g)	
	12weeks	16weeks	12weeks	16weeks
V4S1	274.37 <sup>bc</sup> ±7.92	282.92 <sup>a</sup> ±10.76	32.00 <sup>d</sup> ±1.22	37.79 <sup>c</sup> ±1.37
V4S2	276.05 <sup>bc</sup> ±9.17	278.13 <sup>a</sup> ±10.98	35.66 <sup>c</sup> ±1.75	42.13 <sup>a</sup> ±2.21
V4S3	260.96 <sup>c</sup> ±7.06	275.84 <sup>a</sup> ±7.29	35.24 <sup>c</sup> ±0.19	41.47 <sup>ab</sup> ±0.29
V1S1	265.21 <sup>c</sup> ±4.53	283.54 <sup>a</sup> ±14.12	31.73 <sup>d</sup> ±0.92	33.79 <sup>d</sup> ±0.16
V1S2	310.62 <sup>a</sup> ±15.36	275.21 <sup>a</sup> ±29.61	37.45 <sup>b</sup> ±0.28	39.48 <sup>bc</sup> ±1.27
V1S3	288.13 <sup>b</sup> ±0.63	287.71 <sup>a</sup> ±13.48	39.54 <sup>a</sup> ±0.43	35.30 <sup>d</sup> ±1.52

Means with different superscript (abcd) within the same column are significantly different ( $p < 0.05$ ). Values are means  $\pm$  standard deviation of three replicate. Samples code: V4S1–Ex-igbariam 20cm, V4S2–Ex-igbariam 30cm, V4S3–Ex-igbariam 40cm, V1S1–Umuspo 3 20cm, V1S2–Umuspo 3 30cm, V1S3–Umuspo 3 40cm

Table 3 show the flavonoid content of the OFSP varieties. Umuspo3 V1S2 (spaced at 30cm) had the highest flavonoid content (310.62mg/100g) while Ex-Igbariam V4S3 (spaced at 40cm) had the least value (260.96mg/100g) at 12<sup>th</sup> week of harvest. Ex-Igbariam V4S1 and V4S2, V4S3 and Umuspo 3 V1S1 were statistically similar ( $p > 0.05$ ). At 16<sup>th</sup> week of harvest, no significant difference was observed in the flavonoids content of Umuspo 3 and Ex-Igbariam irrespective of the planting spaces. Umuspo3 samples planted 30cm and 40cm apart had higher flavonoid content than Ex-igbariam samples planted in the same spacing, while Ex-Igbariam V4S1 planted at 20cm apart in row had higher flavonoid value than Umuspo-3 V1S1 planted 20cm apart in row.

Polyphenol content of the OFSP varieties are also shown in Table 2. Umuspo 3 V1S3 had the highest value of 39.54mg/100g at 12<sup>th</sup> week. Ex-Igbariam V4S2 and V4S3 were statistically similar ( $p > 0.05$ ), likewise V4S1 and V1S1. At 16<sup>th</sup> week, V4S2 had the highest total polyphenol content (42.13mg/100g) while V1S1 had the least value (33.79mg/100g). There was an increase in the total polyphenol content at 16<sup>th</sup> week except for V1S3 which decreased by 11.55%. It was observed that 20cm planting space had higher value of polyphenol for both varieties.

## Conclusions

This study has shown the effects of planting distance and harvesting period on the phytochemical and carotenoid concentration of OFSP varieties. We found that Umuspo 3 and Ex-Igbariam varieties contained significant amount of phytochemicals and carotenoids. It was observed



that the carotenoid profile of Umuspo 3 variety had highest concentration and Vitamin A (Retinol equivalent) than that of Ex-Igbariam. Furthermore, Umuspo 3 peel and flesh samples planted 30cm and harvested 12<sup>th</sup> week had the highest carotenoids concentration. This article has provided the evidence that planting space and harvesting periods promoted phytochemical and carotenoids accumulation in the OFSP varieties. Umuspo 3 OFSP variety could be planted at 30cm and harvested at 12<sup>th</sup> week, and Ex-Igbariam planted 30cm and harvested at 16<sup>th</sup> week respectively for optimal phytochemical and carotenoids retention. When OFSP varieties planted on the utisol of South East Nigeria are harvested after 12 weeks, the peel will contain more carotenoids than the flesh. The carotenoid distribution becomes more balanced between the flesh and the peel by the 16<sup>th</sup> week.

### Conflict of Interest

Authors declares no conflict of interest.

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