



COMPARATIVE EVALUATION OF PHYSIOLOGICAL POST-HARVEST ROOT DETERIORATION, TOTAL CAROTENOIDS, STARCH CONTENT AND DRY MATTER OF SELECTED CASSAVA CULTIVARS

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Abstract

Cassava root storage is limited by Post harvest Physiological deterioration (PPD), which renders cassava root unpalatable and unmarketable. The research work was aimed at analyzing delayed PPD, total carotenoids (TC), starch content (SC) and dry matter (DM) of some selected cassava cultivars and correlating the variables to ascertain their relationships. We planted twenty cassava cultivars at the western farm of the National Root Crop Research Institute (NRCRI), Umudike which were replicated twice and the different cultivars served as the source of variation. The design of the experiment was Randomized Complete Block Design (RCBD). The cassava cultivars were harvested after the twelfth month of planting and root and shoot biomass taken. The roots obtained were taken to the laboratory to evaluate for PPD, TC, SC and DM. The results obtained were analysed using GENSTAT to obtain the ANOVA, Correlation Coefficient and the mean separated using LSD. The correlation result showed that PPD was inversely correlated to TC, although the association was weak. SC and DM were however, directly correlated to PPD. TC was inversely correlated to SC and DM, although their correlation was weak. SC and DM were directly and strongly correlated.

Keywords: Post-harvest deterioration, Total carotenoids, Dry matter

Introduction

Cassava production and marketing has been limited by its response to abiotic stress known as Post Harvest Physiological Deterioration (PPD) which renders cassava unpalatable and unmarketable (Zainuddin *et al.*, 2017). This deterioration occurs in healthy roots kept in the storage, thus may not be associated with diseases and pests (Reilly *et al.*, 2004). A serious constraint to cassava production is the short shelf life of its roots due to post-harvest physiological deterioration (PPD). PPD begins within 24 hours, and rapidly renders the roots unpalatable and unmarketable. Consequently, cassava roots need to be consumed soon after harvesting. The short shelf-life severely limits the marketing options because it increases the likelihood of losses, marketing costs, and access to urban markets is limited to those close to the production sites (Reilly *et al. ibid*).

Cassava (*Manihot esculenta* Crantz) together with some other crops like maize (*Zea mays*), yam (*Dioscorea spp*) form an important staple food for sub-Saharan African countries (Allen, 2002). For 750 million people around the world, cassava is one of the most important sources of dietary carbohydrates with its starchy root being the most harvested (Allen, 2002; Gleadow *et al.*, 2009; Burns *et al.*, 2011). The genus *Manihot* comprises 98 species spread throughout the Neotropics, 17 of which

are native to North America and the others to South America (Rogers and Appan, 1973). Cassava is one of the most efficient producers of edible carbohydrates among all the world's major food crops when cultivated under optimal conditions which give good yield. In spite of the nutritional deficiencies, cassava remains a key source of calories to the poverty stricken people in the world; this is linked to its agronomic properties. Cassava grows well under marginal conditions in degraded and acidic soils with minimal technical effort and has a flexible harvesting time ranging from 8 – 24 month after planting. Cassava is also amenable to piece meal harvest, which is the harvest of a couple of roots from a plant as needed, while the rest of the roots stay buried underground until harvest.

A large proportion of cassava accessions are drought tolerant and also resistant to pest and diseases (due to the presence of cyanogenic glycosides). These attributes make cassava an attractive crop for small-scale farmers with limited resources particularly in sub-Saharan African populations (Ceballos *et al.*, 2004; Wenham, 1995). García and Dale (1999) and Nyerhovmo (2004) reported that the world cassava production has increased in recent years and is expected to increase further primarily due to its higher demands as human food and industrial value, mostly in Africa. However, Wenham

(1995) noted that the expansion of this crop is drastically restricted by the storage potentials of the roots, which is limited to only a few days. Beeching *et al.* (2002) reported that the short storage life of cassava roots is directly linked to an endogenous physiological process known as physiological postharvest deterioration (PPD), which is considered to be a complex procedure linked to enzymatic stress response to wound. Within 48 hours of harvesting, the roots of cassava suffer an abiotic stress-response known as post-harvest physiological deterioration (PPD) (Beeching *et al.*, 1998). This response renders the root unpalatable and unmarketable. With increasing distance between farmers and markets due to urbanization, PPD has become a major constraint to the development of cassava for farmers, processors and consumers.

PPD is a response of the cassava root to the wounds induced during the harvesting process and is not due to microbial action, although microbial deterioration can set in subsequently (Booth, 1976; Noon and Booth, 1977). Visually, PPD is observed as a blue fluorescence under UV light and a blue-black streaking of the vascular tissues. In addition, coloured occlusions and tyloses from the adjacent parenchyma are seen to block xylem vessels (Richard *et al.*, 1979). A range of secondary metabolic products accumulate during PPD, including hydroxycoumarins, flavan-3-ols, lipids, steroids and diterpenoids (Lalaguna and Agudo, 1989; Richard, 1981). Hirose (1986) and Uritani *et al.* (1984) observed that during Post-harvest physiological deterioration (PPD), the cassava root is biochemically active. PPD is accompanied by increases in respiration in the root and some mobilization of starch into sugars. The objectives of the study are to- determine total carotene content and analyse some growth parameters of the 20 cassava cultivars like stem girth, plant height and plant canopy; determine the harvest index and root biomass, assess the rate of post-harvest physiological deterioration among the 20 cultivars in 5days, 10days and 15days; and correlate the impact of total carotene content on Post-harvest physiological deterioration, growth, starch content and dry matter.

Materials and Methods

Sources of Material

Twenty cassava cultivars used for the study were obtained from the Genetic Resource Unit of National Root Crop Research Institute (NRCRI), Umudike in Abia State, Nigeria.

Location of study

The study was carried out at Western Farm of National Root Crop Research Institute (NRCRI), Umudike located at Longitude 07° 24' E and Latitude 06° 52' N of the equator and altitude 122m above sea level.

Experimental design/statistical analysis

The plant samples were planted in the Western Farm of NRCRI, Umudike on the 27th July, 2015. The design of the experiment was Randomized Complete Block Design (RCBD), replicated twice. The different cultivars served as source of variation. The 20 cassava

cultivars planted were harvested on the twelfth month and their shoot and root biomass measured and recorded. After the biomass has been taken, the cassava roots were taken to the lab for beta-carotene extraction, starch extraction, dry matter and post-harvest physiological deterioration analysis. Data generated from the 20 cultivars were subjected to statistical analysis. They are B2-37, COB517, NR110348, TMS011797, NR100248, NR090187, NR090182, B1-5, NR110031, NR060251, B1-29, NR050362, COB427, NR100462, NR110078, NR110165, NR110084, CR52A4, NR100216 and B1-42.

Beta-carotene extraction from cassava root

Apparatus/equipment: sensitive weighing balance, mortar, pestle, distilled water, silicate, acetone, brine, foil, petroleum ether, sodium sulphate anhydrous, filter paper, separating funnel, beakers, retort stands, conical flask, cassava root, kitchen knife, bucket, water.

Procedure

The cassava roots were harvested from the Western Farm in Umudike and each of them were properly labelled for easy identification. The labelled samples were taken to the Beta-carotene laboratory for extraction. The twenty cultivars (cassava roots) were thoroughly washed with clean water and were placed in separate plates, and each plate labelled according to the label on the Sample. The cassava roots were peeled. After each sample has been peeled the knife was washed thoroughly before we peeled another sample. The peeled cassava roots were chopped (i.e. cut into small pieces) using the kitchen knife. The chopped samples were placed in separate plates and labelled properly. Then using the foil, we measured out ten grams (10gms) from each of these plates. Note that the foil was weighed first and then set to zero reading before adding the chopped sample in order to get the weight of the sample only.

Carotenoids were extracted following the method of Safo-Katanga *et al.* (1984). The Beta carotene was poured into a cuvette and the absorbance reading taken using the spectrophotometer.

Spectrophotometer reading and calculation

1. The volume was made up with petroleum ether and the absorbance was taken at 450nm. The carotenoid solution may be concentrated or diluted if necessary. Absorbance should be between 0.2 and 0.8.
2. The total carotenoid content is calculated using the following formulae:

Total carotenoid content (mg/g) =

$$\frac{A_1 \times \text{Volume} \times 10^4}{A_2 \times \text{Sample Weight (g)}} \dots\dots (1)$$

Where,

A₁ = Absorbance

V = Volume

A₂ = Absorption coefficient of B-carotene in PE (Petroleum ether)

Finally, multiply by 100 to give the carotenoid content in mg/100g.

Extraction of starch from white root cassava genotypes

Material/Equipment: Refridgerator, electric blender, oven, beaker, sensitive weighing balance, sieve, moisture can, kitchen knife, bucket, cassava roots, plates and water.

Procedure: The white cassava roots were harvested from NRCRI farm in Umudike and were labelled properly. The samples were taken to the molecular biology laboratory where the cassava roots were properly washed with clean water and placed on a labelled plate (the plates were labelled according to the label on the cassava sample) and then starch extracted from the cassava root using Benesi *et al.* (2004) method.

Dry matter preparation from cassava root tubers

Material/Equipment: Oven, moisture can, kitchen knife, beaker, bucket, water, sensitive weighing balance.

Procedure: Percentage dry matter content (DMC) of the roots was estimated using Benesi *et al.* (2004) method. The 10g of the chopped cassava roots were weighed out from each of the labeled plate in the second experiment using the sensitive weighing balance. Ten gram (10g) of the chopped cassava root were put in a well labelled moisture cans. The weight of the moisture cans were taken before the samples were put into the can and then reweighed after they were put into it. The weighed samples were put into the oven for drying. The samples were left in the oven to dry properly, reweighed and recorded.

The weight of the dry matter (Dm) was estimated thus:

$$D_m = \frac{\text{Weight of can} + \text{dry weight of sample} - \text{weight of empty can}}{\text{Initial weight of can}} \times 100 \dots (2)$$

Moisture content = 100 – Dm

Evaluation of post -harvest physiological deterioration in cassava

Material/Apparatus: PVC film, scissors, stainless steel knife, wooden or plastic steel shelves, table.

Procedure: Commercially sized roots with a minimum length of 18cm, without mechanical damage and with no pre-harvest rotting visible were selected. The proximal and distal root ends were cut and ensured that the remaining root section was at least 15cm long after the

ends were cut. The distal end of the root was covered with PVC film in order to maintain moisture content of the distal end and inhibit the development of deterioration from the end of the root. Physiological deterioration developed only from the proximal end, where loss of tissue moisture occurred. The roots were stored on steel shelves at 21-28°C and 70-80% relative humidity in a place protected from direct sun, rain and rodents, but exposed to air. Root section after three days of storage was evaluated, during this period there was sufficient time for physiological deterioration to develop, but insufficient for microbial deterioration. The evaluation was carried out as follows:

i) Transverse slices of; 2, 4, 6, 8, 10, 12 and 14cm were cut from the proximal end, giving a total of seven slices.

ii) Values from 0% to 100% PPD were assigned based on the extent of physiological deterioration on the surface of each slice of root. Usually, only the periphery of the parenchyma is considered, since the central tissues rarely deteriorate. It helps to divide this part of the root surface into 10 sections and estimate deterioration in each section.

iii) Each of the seven slices; 15, 30, 45, 60, 75 and 90% of the total length were scored using visual inspection according to Wheatley *et al.* (1982) and the mean for the entire root calculated and recorded.

Results and Discussion

Post-harvest physiological deterioration

From the results shown in Table 1, COB517 (0.71) had the least PPD result followed by NR110031 (0.86), NR100248 (1.03) and B2-37 (2.43). Cultivar NR110165 (29.86), B1-42 (26.49) and NR110084 (25.54) had the highest PPD; the lower the PPD, the better the performance. At day ten (10), cultivar B2-37 (3.00) had the least PPD, followed by TMS011797 (5.57), NR100248 (5.86), COB517 (6.07) and NR110348 (6.25). Cultivar B1-42 (61.79), NR100216 (47.43), B1-29 (41.43) and NR050362 (41.07) had the highest. At day fifteen (15), B2-37 (8.80) had the least PPD, followed by COB517 (10.50), TMS011797 (11.30), NR110348 (13.10) and NR100248 (19.00). Also, cultivar B1-42 (72.50) had the highest PPD, followed by NR100216 (62.70), CR52A4 (57.90) and NR110084 (48.80). There were significant differences ($P \leq 0.05$) among the twenty (20) cultivars.

Table 1: Data for cassava Post harvest physiological deterioration for day five, ten and fifteen

S/N	Genotype	PPD DAY5	PPD DAY10	PPD DAY15
1	B2-37	1.71±2.43 ^b	3.00±3.03 ^a	8.80±5.05 ^a
2	COB517	0.71±0.2 ^a	6.07±0.51 ^{bc}	10.50±5.30 ^b
3	NR110348	1.57±2.22 ^b	6.25±3.32 ^c	13.10±1.68 ^b
4	TMS011797	4.96±2.58 ^d	5.57±1.82 ^b	11.30±6.26 ^b
5	NR100248	1.03±.95 ^{ab}	5.86±4.24 ^b	19.00±3.9 ^c
6	NR090187	3.04±2.78 ^c	11.19±1.68 ^d	19.00±7.41 ^c
7	NR090182	3.57±4.65 ^c	6.43±3.03 ^c	24.60±0.40 ^d
8	B1-5	3.62±1.75 ^c	10.46±6.22 ^d	21.00±19.59 ^c
9	NR110031	0.86±1.22 ^a	12.32±4.79 ^d	22.60±2.83 ^{cd}
10	NR060251	2.82±.45 ^{bc}	12.86±7.58 ^e	20.50±6.13 ^c
11	B1-29	2.29±1.21 ^b	41.43±16.16 ^h	54.40±0.1 ^f
12	NR050362	7.43±4.85 ^e	41.07±4.55 ^h	52.10±11.11 ^f
13	COB427	19.43±1.22 ^g	26.43±9.09 ^f	58.00±3.97 ^g
14	NR100462	24.29±10.5 ^h	33.57±21.21 ^g	50.60±2.82 ^e
15	NR110078	16.14±3.84 ^f	29.36±12.22 ^f	63.00±9.86 ^h
16	NR110165	29.86±12.85 ⁱ	32.86±14.14 ^g	48.00±7.91 ^e
17	NR110084	25.54±21.97 ⁱ	38.85±24.64 ^g	48.80±21.06 ^e
18	CR52A4	20.00±12.12 ^g	36.43±9.09 ^g	57.90±16.36 ^g
19	NR100216	25.72±14.14 ⁱ	47.43±2.42 ^h	62.70±16.36 ^h
20	B1-42	26.49±15.74 ⁱ	61.79±28.79 ⁱ	72.50±17.03 ⁱ

Values expressed as Mean± Standard error ($P \leq 0.05$) below the column

Starch content

The starch, beta-carotenoid and dry matter content of twenty (20) cassava cultivars were analyzed and presented in Fig. 1. Results show that cultivar NR110031 (32.1g) had the best performance for starch

content, followed by NR110348 (30.5g), B1-42 (30.0g), NR100216 (29.6g) and NR110165 (29.2g). Among the least were; B2-37 (10.3g), NR060251 (15.2g), NR050362 (16.1g) and NR090187 (16.9g).

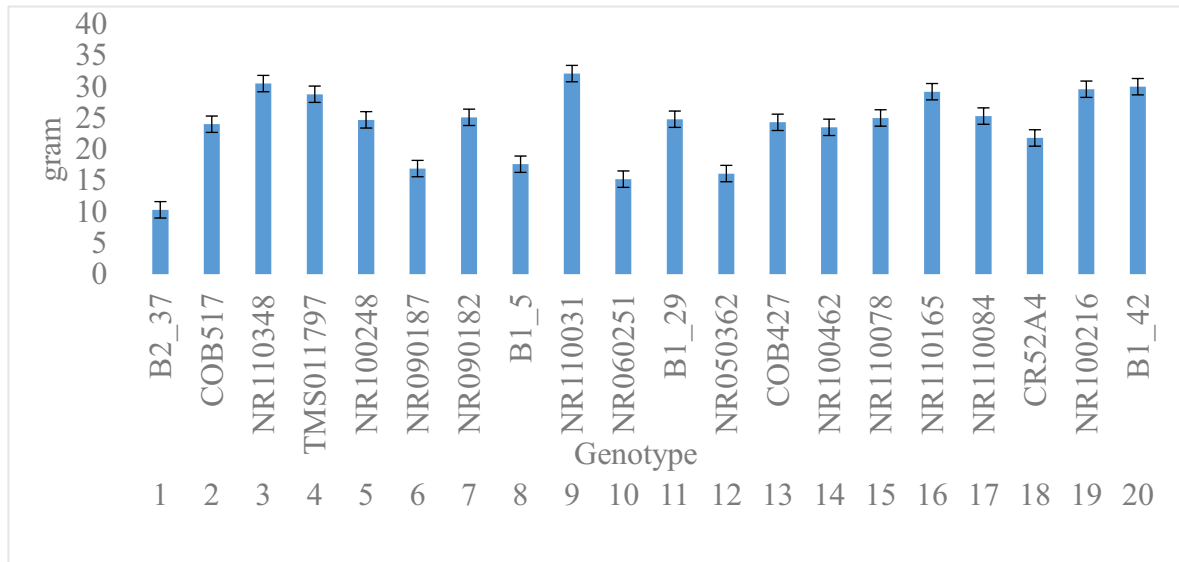


Figure 1: Chart showing the starch content of twenty cassava cultivars measured in grams

Total carotenoid

From the result in Figure 2, cultivar B2-37 (9.01) had the best total carotenoid performance, followed by NR090187 (8.63), NR060251 (4.57) and B1-42 (3.63). Among the least were; NR050362 (0.82g), NR100216 (0.87) and COB427 (1.03).

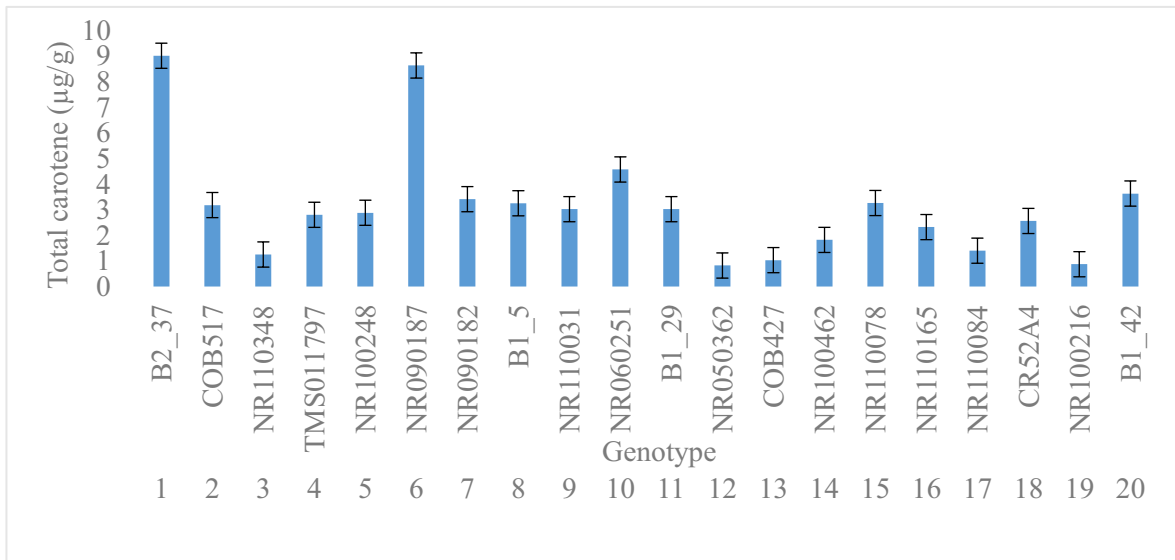


Figure 2: Chart showing total carotenoids of twenty cassava cultivars

Dry matter content

The result for dry matter of the twenty (20) roots of the cassava cultivars is shown in Figure 3. Cultivar NR110348 (46.65g) had the highest dry matter content,

followed by TMS011797 (43.07g), CR52A4 (42.50g), B1-42 (42.00g) and B1-29 (40.80). The least were; B2-37 (24.00g), NR060251 (25.50g), NR050362 (27.85g) and B1-5 (29.00g).

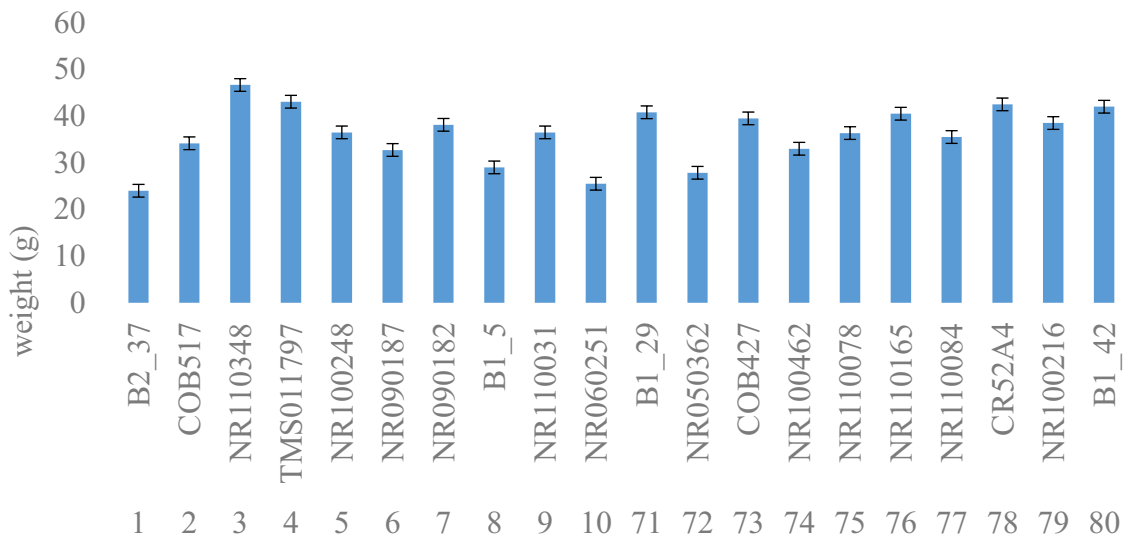


Figure 3: Chart showing the dry matter content of twenty cassava cultivars measured in grams

Correlation between PPD, total carotenoids, starch content and dry matter

Result in Table 2 shows the correlation between PPD, total carotenoids, starch content and dry matter of twenty (20) cassava cultivars. PPD at day five (5) inversely correlated with total carotenoids, but the correlation was weak. PPD at day ten (10) and day fifteen (15) inversely correlated with total carotenoids, but at day 10, showed weak correlation and day 15, very weak correlation. This observation shows that as the value total carotenoids increase, the PPD improved. At day 5, 10 and 15, PPD had direct correlation with starch

content and dry matter each. This implies that as the value of starch content and dry matter increases, PPD decreased. However, the correlation at day 5 was weak; while at day 10 and 15, it was very weak. Results also show that total carotenoids and starch content was inversely correlated, but weak. Thus an increase in total carotenoids in the cassava root will result to a decrease in the cassava root starch content. There was also a direct correlation between starch content and the dry matter of the cassava root. The correlation between starch content and dry matter was moderate.

Table 2: Correlation between PPD, total carotenoid, starch content and dry matter

Rows	PPD Day 5	PPD Day 10	PPD Day 15	Total Carotenoids	Starch Content	Dry Matter
PPD Day 5	1					
PPD Day 10	0.608*	1				
PPD Day 15	0.469*	0.688*	1			
Total	-0.267 ^{ns}	-0.224 ^{ns}	-0.161 ^{ns}	1		
Carotenoids	0.211 ^{ns}	0.194 ^{ns}	0.061 ^{ns}	-0.363*	1	
Starch Content	0.144 ^{ns}	0.199 ^{ns}	0.118 ^{ns}	-0.18 ^{ns}	0.699*	1
Dry Matter						

*PPD=Post-harvest physiological deterioration, *= significant, ns= not significant*

Cassava production and storage has been limited by PPD. From the result (Table 2), dry matter directly correlated to PPD, although, the correlation was weak. This finding conformed to the work done by Cortes *et al.* (2002) and Chavez *et al.* (2005). Unlike Oirschot *et al.* (2000), we found that PPD positively correlated with the dry matter content in the roots, although association (not significant) was weak ($r = 0.144, 0.199$ and 0.118) at day 5, 10 and 15 respectively. This observation is also in disagreement with the report of Sanchez *et al.* (2006), who reported that the association was significant. Gloria and Uritani (1984) reported that a low β -carotene in the root tissues of cassava hastened the PPD. Chavez *et al.* (2005), reported that carotenoid content in the root above 5mg per kg fresh weight delayed the development of PPD symptoms. Carotenoids have been related in the literature to delay or reduce postharvest deterioration in cassava roots (Morante *et al.*, 2010, Sánchez *et al.*, 2006). Variations in degree of development and severity of PPD among the cassava genotypes and within the same genotype have been reported (Buschmann *et al.*, 2000). Kawano and Rojanaridpiched (1983) reported that PPD was affected by the environment where the plants were grown.

Oirschot *et al.* (2000) reported that PPD was positively and significantly correlated with the dry matter content in the roots. Thus an increase in dry matter will result to an increase in PPD. Which implies that; to improve delayed PPD in cassava roots, the dry matter content of such root should be reduced. This observation is disheartening, although the correlation was not significant. The highest dry matter content recorded in this evaluation was lower (46.65%) than that reported by Rajendran and Hrishi (1982), as 66.4% and Magoon *et al.* (1973), as 47.2%. Also, starch content is directly correlated to PPD. The average values for PPD and dry matter content were in accordance with those observed in other studies (Cortex *et al.*, 2002; Chavez *et al.*, 2005). However, from our observation, an increase in starch content will result to an increase in PPD (0.211, 0.194 and 0.06 for 5, 10 and 15 days respectively). Thus, to improve cassava root PPD performance, the starch content of the cassava root should be reduced. However, there was a weak negative correlation between starch content and PPD in the cultivars studied. Oirschot *et al.* (2000) reported negative correlation between PPD and sugar/starch ratio, in contrast with Wheatley and Gomez (1985), who found no correlation between PPD and starch content. An important objective in cassava breeding programs is to increase root dry matter content

because the buyer pays for starch rather than water and a higher dry matter facilitates drying of roots and the extraction of starch. While our finding related to PPD and dry matter content was somewhat discouraging, because in some countries cassava is cultivated for their starch.

The highest total carotenoids recorded in this evaluation (B2-37=9g/g) was higher compared to the observation of Saravanan *et al.* (2015), who reported highest total carotenoids of $0.78\mu\text{g/g}$ in cultivar CI-896. The association between total carotenoid concentration and PPD was negative, although, weak ($-0.267, -0.224$ and -0.161) for day 5, 10 and 15 respectively and not significant. This observation also conforms to the study done by Chavez *et al.* (2005), who reported that correlation between PPD and total carotenoids was negative. However, our observation is in contrast with Chavez *et al.* (2005) who reported a significant association. This observation that an increased beta-carotenoid will result to a decreased PPD, shows that increased beta-carotene helps in increasing shelf life, thus making them more marketable. This may be due to the antioxidant property of the carotenoids. Nevertheless, farmers are encouraged to grow yellow rooted pro-vitamin A cassava cultivars, particularly where cassava is used for human consumption.

The implications of these findings for the genetic improvement of cassava are important. In addition to the nutritional benefits to humans, yellow pro-vitamin A cassava roots are of higher value to the animal feed industry. Carotenoid and dry matter content are independently inherited. A higher dry matter is particularly desirable for the animal feed and starch industries. Among consumers of boiled roots, preference is for varieties that have intermediate levels of dry matter. Market accessibility is specifically a challenge for fresh cassava destined for human consumption as a result of early PPD; consequently any reduction or delay in PPD will be of most benefit to cassava production. However, the starch industry is unlikely to favour yellow cassava because many consumers require a white root product that could not be obtained from yellow roots. The livestock feed industry, in contrast, may benefit from yellow cassava roots because the reduction in PPD may help neutralize the faster deterioration anticipated from the high dry matter roots needed by the industry.

Conclusion

From the twenty cassava cultivars used for this study, B₂-37 and COB517 showed a promising delayed PPD, while B₁-42 and NR100216 were the least. There were also significant differences in the PPD. The result obtained showed that PPD and total carotenoids were indirectly correlated. Though, the correlation was weak and not significant. However, from our observation, there was positive correlation between PPD and starch content and dry matter content. The use of cassava is all encompassing; ranging from food for households to starch for industries. Therefore production of cultivars like B₂-37 and COB517 that are promising with regards to total carotenoids should be encouraged to improve the nutrition and shelf life of cassava. Moreover, cultivars like B₁-42 and NR100216 that have high starch content should also be recommended to farmers for industrial purposes. Furthermore, cultivars like B₂-37 and COB517 with higher total carotenoids should be released to rural farmers for cultivation. Rural farmers should be enlightened on the health benefits of carotene and should be encouraged by making such cultivars readily available to them.

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