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Effect of Post-harvest Handling Conditions on Polyphenol Content and Ferric Reducing Antioxidant Properties of Selected Ugandan Sweet Potato Varieties

Agnes Nabubuya^{a, *}, Yusuf Byaruhanga^a, Ali Mwakha^a, Judith Narvhus^b and Trude Wicklund^b

^a Department of Food Technology and Nutrition, School of Food Technology, Nutrition and Bio-engineering, Makerere University, P.O. Box 7062, Kampala, Uganda
 ^bFaculty of Chemistry, Biotechnology and Food Science, Norwegian University of Life Sciences, Postbox 5003, 1432 Aas, Norway

ybbyaru@yahoo.com; <u>mwakhaali@gmail.com</u>; judith.narvhus@nmbu.no; <u>trude.wicklund@gmail.com</u>, *<u>agnesnabubuya@yahoo.co.uk</u>

*Corresponding author

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Abstract

Sweet potatoes are rich in nutrients, including bioactive compounds like carotenoids and polyphenols. This study provided information on the effect of postharvest handling conditions; curing (direct sunshine) and ambient storage on the total carotenoids, total polyphenols and antioxidant activity of five Ugandan sweet potato varieties. The total carotenoid content was determined following Rodriguez and Kimura's method, while total polyphenol content and antioxidant activity were assessed using Folin-Ciocalteu's reagent and the FRAP assay, respectively. Findings showed significant variation in total carotenoids, total polyphenol content and antioxidant activity of the different sweet potato varieties. There was an increase in total carotenoids in both cured (322 to 391 µg/100g) and non-cured roots (325 to 366 µg/100g), on average during storage. The total polyphenol content declined from 1.04 mg GAE/g (at harvest) to (0.42 and 0.47 mg GAE/g) in cured and non-cured roots, respectively, on average. The antioxidant activity was reduced from 8.74 µmol/g (at harvest), to 3.40 and 3.73 µmol/g in cured and non-cured roots respectively, on average. This study concluded that curing under direct sunshine decreases the polyphenols and antioxidant activities in all varieties but increases total carotenoid content, suggesting careful consideration of postharvest handling procedure before storage, for nutrient maximization.

Keywords: Sweet potatoes; Total carotenoids; Post-harvest handling; Antioxidant activity; Curing

Introduction

Sweet potato is an important crop worldwide and is consumed especially in the tropical regions where it is indigenous. Sweet potatoes are considered to be highly nutritious because they are not only rich in energy, but also contain biologically active components such as polyphenols, β -carotene, ascorbic acid and

dietary fiber (CartabianoLeite et al. 2020, Alam 2021, Laveriano-Santos et al. 2022). The flesh colours of sweet potato genotypes consumed by humans are mainly white, cream, yellow, orange and purple. Purplefleshed varieties exhibit higher levels of phenolic compounds and antioxidant capacity compared to orange-fleshed varieties, except for carotenoid (Im et al. 2021, Oloniyo et al.

2021). Although 'white-fleshed' (white. cream, yellow and pale orange) sweet potatoes are the most commonly cultivated varieties for human consumption in sub-Saharan Africa (Alam 2021), promotion of orange-fleshed varieties was in the recent past initiated among sweet potato consumers to improve the vitamin A status especially among children (Girard et al. 2021). The high phenolic and β carotene content of some sweet potato varieties has resulted in sweet potato being regarded as a functional food (Amagloh et al. 2021). Sweet potato polyphenols have been reported to inhibit the growth of several cancers (Vishnu et al. 2019) and ameliorate diabetes in humans (Zhang et al. 2016). Despite the availability of much information concerning the antioxidant properties of sweet potatoes, most of the work has been carried out using predominantly purple and orange-flesh varieties (Padda and Picha 2008, Rautenbach et al. 2010). Some of the results documented indicate differences in phenolic composition and antioxidant activity in of sweet potato varieties in the different portions like roots and leaves (Im et al. 2021, Padda and Picha 2008; Islam et al. 2003, Kourouma et al. 2020). Variations have also been noted in nutritional phenolic compounds composition, antioxidant activity with seasons (Suárez et al. 2020).

The phytochemical content of sweet potatoes and other produce can be affected by postharvest handling procedures and storage conditions such as curing and time, and exposure to light caused tissue biochemical responses (Grace et al. 2014). Unlike other highly perishable produce like fruits and vegetable, sweet potatoes remain consumable for days to months (Grace et al. 2014). In some places sweet potato roots are cured by holding in a properly ventilated facility maintained at about 29 °C with 85-90% relative humidity for 4-7 days (Edmunds et al. 2008). The effects of curing and storage on sweet potato antioxidant activity have been previously investigated, but earlier research focused mainly on the use of controlled curing and storage conditions (Edmunds et al. 2008). In Sub-Saharan African, sweet potato roots are subjected to uncontrolled postharvest handling and storage conditions, including ambient storage and pit storage, which can affect the chemical composition of the roots including phenolic and carotenoid content of the roots (Akoroda and Teri 1998, Mpagalile et al. 2007).

Various researchers have reported varying changes in phytochemical and antioxidant activity in sweet potato roots subjected to different postharvest handling and storage conditions. A study by Selokela et al. (2022) reported a 1.2-1.5 times increase in βcarotenoids upon solar drying sweet potatoes at 20-60 °C. Sun et al. (2019), found a 10 fold increase in the total phenolic content during 30 days of storage at room temperature. Priyadarshani et al. (2007) reported 2 times increase in beta carotene in two weeks during open storage. Despite the previous reports, there is limited information on the antioxidant properties of Ugandan sweet potato varieties and the effect of the postharvest handling methods commonly used. The aim of this study was to provide information on the antioxidant components (total polyphenols and total carotenoids) and antioxidant activity of some Ugandan sweet potato varieties. This study also aimed at evaluating the variation in antioxidant activity of cured and freshly harvested sweet potatoes during ambient storage and determining the storage length for maximizing the different antioxidant components.

Materials and Methods Sweet potato materials

Five sweet potato varieties with varying flesh colours (white, cream, and orange) were used in this study. The sweet potatoes were harvested from three replicate experimental fields at the National Agriculture Crop Resource Research Institute (NACCRI) in Uganda. Mature sweet potato roots from all varieties were harvested and handled in two ways before storage; freshly harvested roots were either stored directly (non-cured) or were cured by spreading under the sun for 4 days (29–31 °C and 63–65% RH) before storage. The roots were stored in ambient/room conditions (23–26 °C and 70–80% RH) for eight weeks. Analysis was done weekly for

changes in total carotenoid content and after every 2 weeks for total polyphenols and antioxidant activity.

Sample preparation

Sound roots (200–250 g) were randomly selected for each sweet potato variety from each of the three replicates to make composite samples for subsequent analyses. Each of the selected roots was washed under running tap water, peeled, and sliced longitudinally into four pieces of which two opposite pieces were then grated uniformly. The grated tissue from the three replicates was combined and mixed thoroughly. Samples for total carotenoid analysis were prepared by homogenizing grated tissue in a mortar. Samples for total polyphenol content and antioxidant activity were prepared by freeze-drying grated root tissue with a vacuum freeze-dryer (DetiAnyou FD-27S, Beijing, China) at -30 °C for 24 hours. Just before analyses, the samples were milled using a laboratory mill (Wondermill, model 70, Korea).

Analyses

Total carotenoid content

The total carotenoid content of the sample was determined according to Rodriguez & Kimura (2004). Five (5) grams of the homogenized sample was thoroughly mixed with 50 mL of cold acetone and the mixture was filtered with suction through a Buchner funnel fitted with glass wool. The acetone extract was added to 40 mL of petroleum ether in a 500 mL separation funnel and distilled water was slowly added to the mixture and allowed to flow along the sides of the funnel. After separation, the aqueous phase (lower phase) was discarded and the petroleum ether (PE) phase was washed 3 times with distilled water to remove any residual acetone. The PE phase was collected into a 50 mL volumetric flask, the solution was passed through a small funnel containing anhydrous sodium sulphate (15 g) to remove residual water and the volume was topped up with PE. The absorbance of the carotenoid solution was taken at 450 nm. The total carotenoid content was recorded in $\mu g/100 g$.

Total polyphenol content (TPC) and antioxidant activity

The sweet potato samples were extracted according to the method used by Dincer et al. (2011). Samples weighing 1 g were placed in 50 mL centrifuge tubes and were extracted using 20 mL aqueous methanol (80%) by heating in a water bath at 80 °C for 10 min, shaking manually for 30 s, and cooling the tubes to room temperature before centrifuging at $4,500 \times g$ for 20 min. The supernatants were transferred to 25 mL volumetric flasks and the volumes were adjusted with extraction solution. The methanol extracts were stored at -20 °C until they were analyzed.

Total polyphenol content

The total polyphenolic content (TPC) was analyzed by the procedure based on the Folin-Ciocalteu's reagent (FCR) as described Volden et al. (2008). The analysis was conducted using a Konelab 30i (Thermo Electron Corp. Vantaa, Finland), an automated equipment in which 20 µL of sample were added to 100 μL of FC reagent (diluted 1:100 with distilled water), mixed and incubated at 37 °C for 60 s prior to addition of 80 µL of 7.5% (w/v) sodium bicarbonate solution. The samples were mixed and incubated at 37 °C for 15 min. The absorbance was read at 765 nm and the results were assessed against a calibration curve of garlic acid. The results were presented as mg Garlic equivalents (GAE)/g(DW).

Ferric reducing antioxidant power (FRAP)

Antioxidant activity in the sweet potato methanol extracts was measured using the ferric reducing antioxidant power (FRAP) assay described (Benzie and Strain, 1996) with some modifications by Volden et al. (2008). The measurements were carried out using a Konelab 30i in which 200 μL of the FRAP reagent (3.0 mM acetate buffer, 10 mM 2,4,6-Tripyridyl-S-triazine (TPTZ) in 40 mMHCl, FeCl $_3$ 6H $_2$ O, ratio 10:1:1) were automatically pipetted separately and mixed in the cuvettes with 8 μL of the sample extract. The mixture was then incubated at 37 °C for 10 min and the absorbance read at 595 nm. Trolox (Vitamin E analogue) was used as a

control. The results were expressed as $\mu \text{mol/g}$ Dry matter.

Statistical analysis

Data for the different parameters were analysed using Minitab (Minitab Inc., State College, PA, USA) version 16. Analysis of variance (ANOVA) using adjusted sums of squares for tests was used to analyse for variation in means of total carotenoids, total polyphenols and antioxidant activity among the five varieties of sweet potatoes (with varying flesh colours) during storage. Tukey's test was used to determine which specific means within the different variables among treatments were different from each other. Means were considered to be significantly different at p < 0.05. Pearson product-moment correlation coefficient tests were done to measures the strength and direction of the relationships between the different antioxidant components within the different sweet potato varieties during storage. This method was selected because the data was continuous. The results are presented as means with respective standard deviations obtained from analyses.

Results and Discussion

Effect of postharvest handling conditions on total carotenoids, total polyphenol content and antioxidant activity of ambient stored sweet potato roots

Total carotenoid content

Significant differences (p < 0.05) were observed in the total carotenoid content of the sweet potato varieties used in the present study. NASPOT 10 and NASPOT 2 displayed the highest and lowest total carotenoids (343.4 and 14.75 μ g/g respectively). The carotenoids were significantly higher in orange-fleshed (NASPOT 9 and NASPOT 10) than in the white/cream-fleshed varieties, by 23-fold (Figure 1). The study by Kourouma et al. (2020) aligns with these findings, showing that orange-fleshed sweet potato varieties contain significantly higher total carotenoids

compared to white/cream-fleshed varieties. Their results revealed a 23-fold difference in carotenoid content among the different flesh colours, mirroring results with NASPOT 9 and NASPOT 10 in this study. This consistency underscores the value of orange-fleshed sweet potatoes as rich sources of carotenoids, particularly β-carotene, crucial for addressing vitamin A deficiency in regions with limited access to nutrients. Orange-fleshed sweet potato varieties are being promoted in Africa to improve the vitamin A status of children because they are bio-fortified with β -carotene (Mutuku & Mwaniki 2019, Bao 2020). The total carotenoids in all the sweet potato roots increased during storage, although they decreased consistently after 3-4 weeks of storage in the orange-fleshed varieties (Figure deep orange-fleshed varieties 1). (NASPOT 9 and NASPOT 10) contained the highest and the white-fleshed variety (NASPOT 2) displayed the lowest carotenoid content consistently during storage. Higher expression of carotenoid biosynthesis genes like LCYB and lower expression or activity of the catabolic gene IbCCD4 could contribute to the exceptional carotenoid retention observed in these varieties after curing and storage. NASPOT 9 and NASPOT 10 sweet potato varieties were genetically engineered to increase carotenoid content, specifically βcarotene (Ngailo et al. 2013). Similarly, over expression of the sweet potato Orange-Insunder (IbOr-Ins) gene in anthocyanin-rich purple-fleshed sweet potato enhanced carotenoid accumulation and stability during post-harvest storage (Park et al. 2015). There were significant differences (p < 0.05) in the carotenoid content between the cured and noncured roots during storage with cured roots displaying higher carotenoid content than the non-cured (Figure 1). These results point to the fact that the enzymes responsible for the biosynthesis carotenoids of remained functioning under postharvest handling and conditions, storage thus facilitating carotenogenesis.

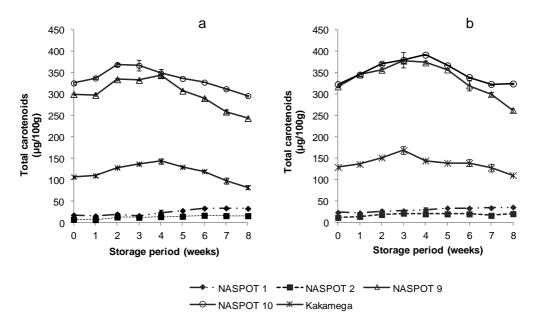


Figure 1: Effect of postharvest handling conditions on the total carotenoid content of five sweet potato varieties stored in ambient conditions (a) freshly harvested (non-cured) roots and (b) cured roots in Uganda.

The findings from this study agree with Selokela et al. (2022) who observed increase in β -carotenoids upon solar drying sweet potatoes at 20-60 °C implying that curing increases carotenoid content in sweet potatoes. Carotenoid synthesis has been reported to have the potential to occur in fruits, vegetables and root crops provided the plant materials are intact, hence preserving the enzyme for carotenogenesis (Simões et al. 2020, Tripathi et al. 2020). The postharvest degradation of carotenoids observed after 3-4 weeks of storage in the orange-fleshed varieties could be attributed to enzymatic or non-enzymatic oxidation (Ngamwonglumlert et al. 2020).

Total polyphenol content

The polyphenol content differed significantly (p < 0.05) among the sweet potato varieties studied with NASPOT 2 and Kakamega having the highest and lowest mean content (1.6 and 0.77 mg GAE/g), respectively. The differences in the polyphenol content among varieties may be attributed to the differences

in quantities and/or types of phenolols synthesized as influenced by genotype (Rumbaoa et al. 2009, Rosero et al. 2020). There was a reduction in the polyphenol content of the sweet potato roots in all the varieties during the 8 weeks of storage at ambient conditions (Figure 2). The mean total polyphenols in freshly harvested roots (1.04 mg GAE/g) reduced to 0.42 and 0.47 mg GAE/g in cured and non-cured roots respectively, in storage. NASPOT 2 and NASPOT 9 contained the highest content of total polyphenols during storage of both cured and non-cured roots consistently (Figure 2). Whereas the polyphenol content of the sweet potato roots decreased significantly during curing, there was no significant difference in the polyphenol content of cured and non-cured roots during storage (Figure 2). These results differ from those in previous studies which reported an increase in sweet potato phenolics during storage. Ishiguro et al. (2007) found that total phenolic content was 1.5 times higher in sweet potato roots stored at 15°C for 4 weeks compared to freshly harvested roots. Similarly, Donado-Pestana et al. (2012) reported that total phenols increased by 20-30% in orange-fleshed sweet potato cultivars after 30 days of storage at 25 °C. Sun et al. (2019) also noted that total phenolic content increased by 1.2-1.5 times in 10 sweet potato varieties during 30 days of storage at room temperature. The authors attributed this increase to the activation of phenylalanine ammonia-lyase (PAL), a key enzyme in the phenylpropanoid pathway, in response to wounding stress during storage (Franková et al. 2022, Gabilondo et al. 2022). The results from this study are however consistent with

Kim et al. (2019) who observed that different forms of heat treatment had a significant reduction effect on Protocatechuic acid and Chlorogenic acid, which are phenolic acids in sweet potato roots. Additionally, Islam et al. (2003) also observed a higher phenolic content (10.1 g/100g) in leaves of sweet potatoes grown at lower temperature (20 °C) than those grown at higher temperature, 30 °C (9.0 g/100g). The reduction in polyphenol content in cured sweet potato root could have been caused by the degradation of the polyphenol component caused by heat (Islam et al. 2003), like temperature and relative humidity.

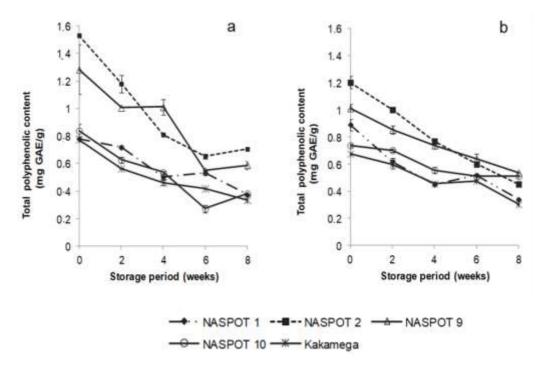


Figure 2: Effect of postharvest handling conditions on the total polyphenol content of five sweet potato varieties stored in ambient conditions (a) freshly harvested (non-cured) roots and (b) cured roots in Uganda.

The differences between results from the present study and the previous studies could be attributed to several factors which include, among other things, differences in sweet potato varieties, environmental conditions, and storage conditions like temperature and relative humidity (Franková et al. 2022, Gabilondo et al. 2022).

Antioxidant activity

There was significant variation (p < 0.05) in antioxidant activity among sweet potato varieties, with NASPOT 2 and NASPOT 9 having higher values (10.4 and 8.45 μ mol/g, respectively) than the other varieties (Figure 3). A reduction in the antioxidant activity was observed in both the cured and non-cured roots

during storage (Figure 3). NASPOT 2 and NASPOT 9 displayed the highest antioxidant activities consistently during the storage of both cured and non-cured roots. The highest antioxidant activity noted in NASPOT 2 and NASPOT 9 sweet potato varieties during storage, both in cured and non-cured roots can be attributed to their higher total polyphenol content (Figure 2). The study revealed a strong positive correlation (p < 0.05, r = 0.815) between antioxidant activity and total polyphenol content in sweet potato roots during storage, indicating that the antioxidant properties are closely linked to the phenolic compounds present.

The sweet potato varieties with the lowest antioxidant activity were observed to have reduced levels of total polyphenol content, leading to diminished antioxidant capacity. The study highlighted that the antioxidant activity decreased in cured and non-cured roots during storage. Varieties with lower antioxidant activities may have lower levels of phenolic compounds, impacting their overall antioxidant potential. Curing caused a reduction in FRAP values in all the sweet potato roots similar to that observed in total polyphenol content (Figure 2b and 3b) but did not significantly affect the FRAP values in the subsequent weeks. The significant positive correlation (p < 0.05, r = 0.815) between antioxidant activity and total polyphenol content of the sweet potato roots during storage shows that as polyphenol content decreased, the antioxidant activities in the sweet potato roots also decreased. This implies a relationship between antioxidant activity and total polyphenol content during storage. These results are in agreement with results from a study by Teow et al. (2007) who found variation in antioxidant activity among sweet varieties and also observed a reduction antioxidant activity during storage, consistent with the decrease noted in both cured and non-cured sweet potato roots over time in this study. The study is also in agreement with Kim et al. (2019) who reported correlation between total phenolic acid content (TPAC), and antioxidant activity of sweet potato according to cultivar and heat treatment condition. However, these results are in disagreement with those reported by Jia et al. (2022) who recorded a negative correlation between total phenols antioxidant activity of sweet potato extracts from the leafy parts of the sweet potatoes. In their study, the cultivar with the highest total phenolic content (5.2 mg GAE/g) exhibited the lowest antioxidant activity (2.8 µmol TE/g), while the cultivar with the lowest phenolic content (3.8 mg GAE/g) had the highest antioxidant activity (4.5 umol TE/g). This could be due to the different plant parts employed in these two studies.

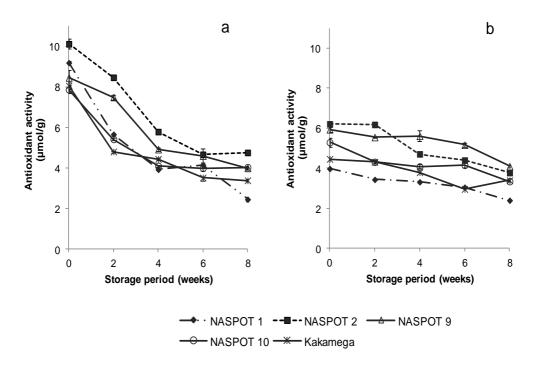


Figure 3: Effect of postharvest handling conditions on the antioxidant activity of five sweet potato varieties stored in ambient conditions (a) freshly harvested (non-cured) roots and (b) cured roots in Uganda.

The present results seemed to suggest that the intensity of orange flesh colour did not translate into high levels of antioxidant activity of the sweet potatoes. This assertion is supported by the findings that whereas orangefleshed varieties (NASPOT 9 and NASPOT 10) in this study consistently had higher total carotenoid content than white and creamfleshed varieties, white-fleshed varieties (NASPOT 1 and NASPOT 2) exhibited higher total polyphenol content and antioxidant activity than some orange-fleshed varieties used in this study except NASPOT 9. There was a significant negative correlation (p < 0.05, r = -0.280) between antioxidant activity and total carotenoid content of the sweet potato roots during storage. This suggests that the color intensity of sweet potato flesh, particularly orange color, is indicative of carotenoid levels rather than total polyphenol content or antioxidant activity. These results are also supported by the significant negative correlation (p < 0.05, r = -0.417) which was

observed between total polyphenol content and the total carotenoid content. These findings are consistent with results from a previous study which showed that carotenoids especially β-carotene do not have ferric reducing activity (Dincer et al. 2011). Toew et al. (2007) also reported poor correlation $(R^2 = 0.480)$ between antioxidant activity the lipophilic-ORAC with the β -carotene contents of sweet potato varieties. The negative correlation between the total carotenoid content and antioxidant activity of the sweet potatoes shows that most of the antioxidant activity of these roots is as a result of the presence of polyphenols (Al-Saikhan et al. 1995, Kalt 2005). However, a study by Kourouma et al. (2019) showed positive correlations between carotenoids antioxidant activity in sweet potatoes. Work on cultivars of berries also showed correlations between carotenoids antioxidant activity Kruczek et al. 2012, Fidrianny et al. 2018). The differences

observed between the present results and the previous work could be due to differences in the food materials and method used to determine the antioxidant activity.

Conclusion

The available evidence from this study indicates that the sweet potato varieties displayed a wide range of antioxidant activity, which was strongly correlated with their total polyphenol content. Specifically, the whitefleshed varieties exhibited the highest antioxidant activity and total polyphenol content. In contrast, the orange and yellow varieties had higher carotenoid content but did not necessarily have the highest antioxidant activity. The study found a significant positive correlation between antioxidant activity and total polyphenol content in sweet potato roots during storage, suggesting that the decrease in antioxidant activities was linked to the reduction in root polyphenol content. Curing and storage led to a decrease in total polyphenol content and antioxidant activity, but an increase in total carotenoid content. There were negative correlations between; total carotenoids and total polyphenol content and between total carotenoids and FRAP values. The authors recommend that to maximize antioxidant activity, sweet potato roots should be consumed while fresh or stored for a short time but should be cured and stored for about 4 weeks to maximize total carotenoid content.

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Conflict of interest

The authors declare that there is no conflict of interest

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