

COMPARATIVE ANATOMICAL EVIDENCE OF THE EFFECTS OF TWO PEELING METHODS ON CASSAVA (*MANIHOT ESCULENTA CRANTZ*) ROOTS

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Abstract

Cassava (Manihot esculenta Crantz) is an important widely cultivated crop in the tropics. The root is encased in a detachable peel made of two layers of varying thickness depending on variety. They have different morphological forms and this makes their peeling by mechanical means difficult and manual peeling is slow and labourious. The effects of chemical peeling methods using NaOH/Citric acid and NaOH/NaCl solutions on the root cells were investigated with microscopy. Elongation and expansion of parenchyma and vessel members of the xylem were apparent in the chemically peeled samples. Conversely, the materials peeled manually showed the presence of tyloses (starch storage structures) and the parenchyma cells were comparatively short. Mean vessel pore diameter was 83.2µm in the manually peeled samples but it was 146.3µm in samples peeled with NaOH/Citric acid and 158.0µm in the samples peeled with NaOH/NaCl solution (p<0.05). The peeling action of the two peeling agent resulted in the disrupted of the integrity of the root cells.

KEYWORDS: Anatomy, Cassava, Chemical Peeling, Manual Peeling

Introduction

Cassava (*Manihot esculenta* Crantz) is a widely cultivated crop in the tropical and sub-tropical regions of the world. In terms of starch content, it is next to corn / maize (*Zea mays*), rice (*Oryza sativa*) and sugar (*Saccharum officinarum*). It is an important component of animal feeds and minimal labour and low financial input are required for its cultivation (Babalaye, 1996; Balagopalan, 2002; Santana *et al.*, 2006; Montagnac *et al.*, 2009). The tuberous roots serve as the reservoir for starch while the fibrous ones help the plant to fix and absorb nutrients and water. The latter root

type is the true root whose ability to absorb nutrients and water decreases considerably with age (Alves, 2002).

The tuberous roots comprise three distinct tissues; Periderm, Peel or cortex and Parenchyma. The Periderm which sloughs off as the root grows old is a thin layer of a few cells which occupies about 3% of the total root weight. The Peel layer or cortex is made up of sclerenchyma, cortical parenchyma and phloem cells; and it occupies about 11-20% of root weight (Barrios and Bressani, 1967). Parenchyma is the starch reserve tissue which comprises

approximately 85% of the total weight of the root. Tyloses are balloon-like structures found in the cell lumen, for storing starch (Pandey, 2002); and they may be present or absent in cassava (Bomfim et al., 2011). In addition, the root xylem has vessels which are radially distributed in a matrix of starch containing cells (Indira and Kurian, 1977; Wheatley and Chuzel, 1993; **Priscila et al., 2013**). The shape and size of cassava root vary consistently within and between populations; and highest variability in size has been recorded in its cultivars than any other known root crops (Wheatley and Chuzel, 1993).

In Africa, a substantial proportion of the food reserve is lost during peeling, using the manual (knife) method. The approach is also time consuming. The alternative peeling method by mechanical means is also not effective or efficient (Akintunde and Oyewale, 2005; Atere et al., 2006; Okigbo, 1980; Sreenarayanan et al., 1995; Ebunilo et al., 2013).

Peeling significantly reduces the cyanide content in cassava. This lethal chemical varies with varieties, within populations and along the root length (Tewe and Iyayi, 1989; Sundaresan et al., 1987; Cooke, 1978; Bradbury et al., 1994; Bokanga and Otoo, 1994; Siritunga and Sayre, 2003) Cyanide must be removed from cassava because it affects man negatively (Halstrom and Moller, 1945; Arguedas and Cooke, 1982; Massaquoi et al., 1990; Osuntokun, 1994; Ernesto et al., 2002a, b; Delange et al., 1994; FAO, 1998, Oluwole et al., 2000; Teles, 2002;

FSANZ, 2005; Wobeto et al., 2007; **Montagnac, et al., 2009**).

This study, therefore, focuses on chemical peeling cassava roots by (i) presenting the effects of the chemicals on the integrity of the root cells, especially the secondary xylem tissue and (ii) documenting changes in morphology of the root cells of chemically treated samples compared with manually peeled ones. The major advantage of peeling by the chemical method is that the entire root is peeled uniformly and there is less risk likelihood of over peeling or under peeling.

Materials And Methods

Cassava roots (TMS 30572) obtained at the International Institute of Tropical Agriculture, (IITA) Ibadan, Nigeria were used for the study. After harvesting, the roots were washed in water and allowed to drain. The procedure of FAO (1977) was adopted for manual peeling of the roots. The approach involved longitudinal cutting up to the cortex using a sharp stainless steel knife to leave the inner white core behind. For chemical peeling, modified methods of Butler and Rivera (2004) and Das and Barringer (2006) were adopted. One batch of the roots was treated with 10% NaOH (1:2, w/v) at 96 °C for 5 minutes and then 3% Citric acid at ambient temperature for 1 minute. The second batch of the roots was separately treated with 10% NaOH (1:2, w/v) at 96 °C for 5 minutes and then followed by 15% NaCl at 96 °C for 3 minutes. The treatments were carried out in a uniscope SM101 water bath. After

treatment, the roots were thoroughly washed in water in order to remove the peels.

Some thin sections of the root tissue were obtained with a sharp razor blade from the basal part (i.e. the middle third portion of the root) which represents an anatomically mature zone of cassava root (Lowe et al., 1982; Appezzato-da-Gloria and Carmello-Guerreiro, 2003). The thin sections were kept in slides containing water. Some samples were placed on slides with few drops of alcohol added. Staining was done with 1-2 drops of iodine solution and thereafter one drop of safranin O. Cover slips were placed on the slides and the edges of the cover slips were ringed with transparent nail varnish to prevent dehydration. Twenty-five different cells of the vessels and parenchyma were examined from four separate fields of the microscope. The features of the roots were captured using a digital camera attached to a CE Olympus compound microscope. The photomicrographs were processed further using CS2 Adobe Photoshop for clarity.

Result

The roots had stratified periderm with some streaks. The parenchyma cells were 3-5 layered in all the samples. In manually peeled samples, the parenchyma cell shape varied from square to more or less rectangular (Fig. 1a, b) whereas, the shape was rhomboidal to imperfectly rectangular in the chemically peeled samples (Figures 2a,b and 3a,b). Starch granules (tyloses) were present in the xylem vessels of

manually peeled samples (Fig. 1a), whereas, they were absent i.e. destroyed as a result of chemical peeling in other samples (Fig. 2a, 3a). The shape of the vessels was circular in the manually peeled samples. The chemicals caused an uneven curvature of the vessel elements. The shape of the vessels varied from uneven square in the samples peeled with NaOH/Citric acid solution to imperfect rectangles in the samples peeled with NaOH/NaCl solution. Tiny clusters of starch granules were observed in the pith area of the samples peeled with NaOH/Citric acid solution (Fig. 3b). Quantitatively, the parenchyma cells were longer in the chemically peeled samples (28.13 μm for NaOH/Citric acid and 30.38 μm for NaOH/NaCl) than the manually peeled specimens (16.83 μm) (Table 1 and Figure 3b, 2b, 1b). Narrower cells were recorded in samples peeled with chemicals than the samples peeled manually (Fig. 1, 2, and 3). Mean vessel pore diameter was 83.2 μm in the manually peeled samples; whereas, it varied from 146.3 μm in the NaOH/Citric acid peeled samples to 158.0 μm in the NaOH/NaCl peeled root samples (Figure 1, 2, and 3 and Table 1) ($p < 0.05$)

Discussion

The different morphological states of the roots of cassava have posed great concerns for engineers (Okigbo, 1980; Sreenarayanan et al., 1995; Egunilo et al., 2013), and has led to failure in fabrication of effective mechanical peeling machines. Where mechanical peelers have been employed, produce loss as result of over

peeling or under peeling results. Therefore, the potential of NaOH/Citric acid and NaOH/NaCl solutions as peeling agents is justified here. The uneven and imperfect rectangular shapes found in the chemically treated samples of cassava roots differed from the perfect circular cells recorded in the manually peeled samples and the isodiametric shape recorded in the two Brazilian accessions reported by Bomfim et al. (2011). Richter and Dallwitz (2009) classified cassava vessels diameter in the medium size class as between 50 μ m and 100 μ m. The value obtained for the manually peeled samples falls within this range whereas the vessel diameter is longer in the chemically peeled samples (146.3 μ m - 158.0 μ m). This observation shows that the chemicals used for peeling caused an expansion of the vessels. Furthermore, the cell area of parenchyma cells was larger in the chemically peeled samples than in the manually peeled ($p < 0.05$). NaOH/Citric acid solution exerted breadth-wise swelling pressure on the vessels while NaOH/NaCl solution caused length-wise extension pressure on the cells. The formation of tyloses has been reported to increase under water deficit conditions and indicates better sap flow in xylem (Rickard and Gahan, 1983; Bomfim et al., 2011), however, our observation did not correlate with this as samples chemically peeled did not have tyloses, this may have been because the chemical peeling treatment may have led to osmotic dehydration of the cells. The results of the study showed that the integrity of root cells of the cassava was affected by NaOH/Citric acid and

NaOH/NaCl solutions. The treatment caused rapid stripping of the periderm and cortex and an elongation as well as extension of the cells ($p < 0.05$). Therefore, chemical method of cassava peeling can serve as an efficient method of removing the periderm and cortex of cassava roots and it can also facilitate a quick release of the stored starch from the roots.

Conclusion

The chemical peeling methods are suitable for removing the peels (periderm and cortex) of cassava roots. The peeling agents namely, NaOH/Citric acid and NaOH/NaCl exerted extension and expansion pressures on the root cells of cassava.

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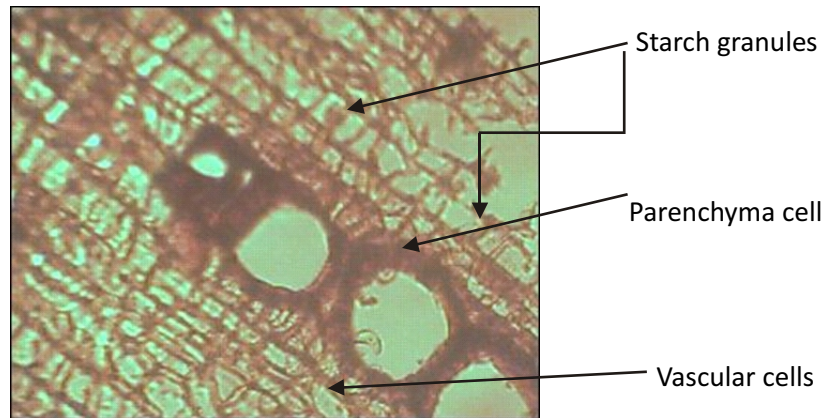


Figure 1a: Micro-Structure of Cassava Root Showing the Vascular Bundle

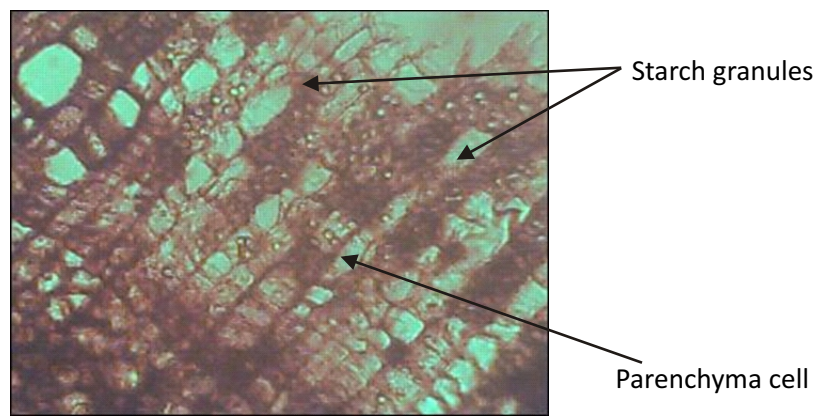


Figure 1b: Micro-Structure of Cassava Root Showing the Parenchyma Cells

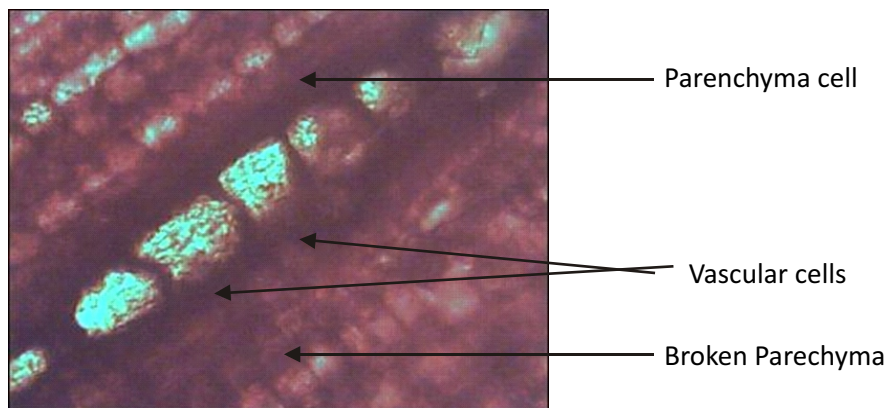


Figure 2a: Micro-Structure of NaOH/NaCl Peeled Cassava Root Showing the Vascular Bundle

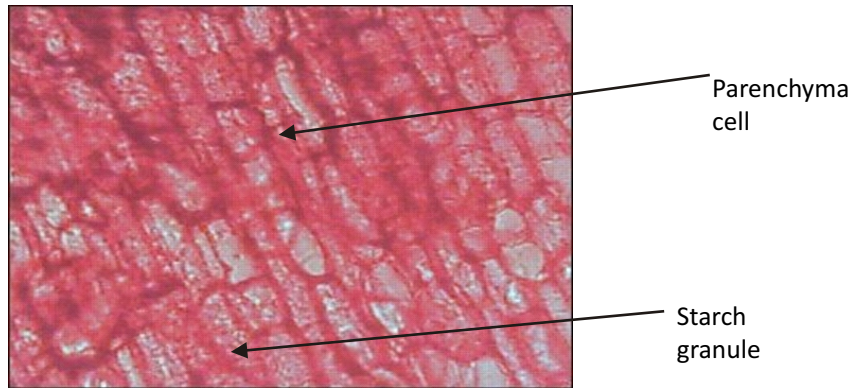


Figure 2b:: Micro-Structure of NaOH/NaCl Peeled Cassava Root Showing the Parenchyma Cells

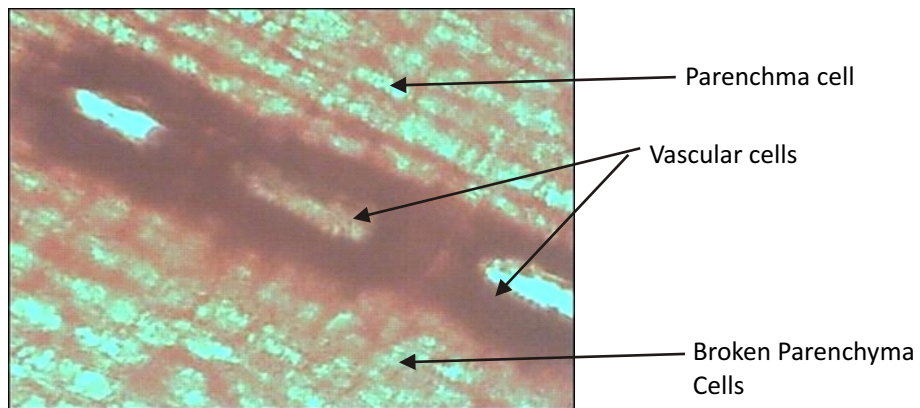


Figure 3a: Micro-Structure of NaOH/Citric Acid Peeled Cassava Root Showing the Vascular Bundle

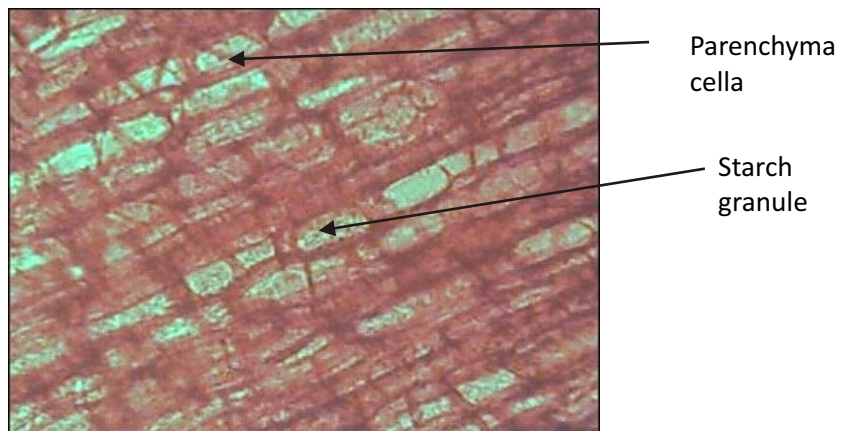


Figure 3b: Micro-Structure of NaOH/Citric Acid Peeled Cassava Root Showing Parenchyma Cells

Table 1: Effects of Chemical Peeling Treatment on Cassava Roots Microstructure

Treatments	P.L (μm)	P.W (μm)	V.L (μm)	V.W (μm)	V.D(μm)
Untreated	16.83 ^a	7.20 ^a	13.00 ^a	13.00 ^a	83.20
NaOH/NaCl	30.38 ^a	7.43 ^a	22.05 ^{ab}	10.13 ^a	158.00
NaOH/citric acid	28.13 ^a	6.30 ^a	25.88 ^b	12.38 ^a	146.30

Note: Cell measured at X400 magnification

Note

- P.L: Parenchyma cell length
- P.W: Parenchyma cell width
- V.L: Vascular cell length
- V.W: Vascular cell width
- V.D: Vascular cell diameter