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Textural and Biochemical Changes Associated with the Hardening Phenomenon in Aiele Fruit (*Canarium schweinfurthii* Eng)

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Abstract: Studies were conducted on the textural and biochemical changes occurring in *Canarium schweinfurthii* Eng fruits, after harvest, in an attempt to study the physical and chemical changes associated with the raw, stored and heat treated samples. The fruits were harvested and stored for 7 days (Stored Hardened fruits), heat treated at 45 °C for 40 min (Heat Softened fruits) and heat treated at 70 °C for 40 min (Heat Hardened fruit). The samples were evaluated for their textural properties, moisture, reducing sugars, total soluble sugars, ash, protein, starch, cellulose, lignin and uronic acids. The hardness of the fruits increased in with storage (15.83 to 20.71 N respectively for raw fruits and stored hardened fruits), while the shear force vary from 0.77 to 5.41 N (respectively for heat softened fruits and heat hardened fruits), making the consumption of heat hardened fruits impossible. Chemical analysis showed no significant differences ($P < 0.05$) between the values obtained for ash, protein, minerals (Na, K, Mg). The moisture of the pulp decreased from 49.32 to 39.90% in a period of 7 days of storage, suggesting that there was dehydration after harvest. Uronic acid, cellulose and lignin increased during storage. Heat hardened pulp showed different changes in the chemical indices than those stored at 22 °C for 7 days, suggesting another mechanism of hardening.

Key words: Canarium schweinfurthii, Hardened, Raw, Doftened, Biochemical, Textural changes

Introduction

Aiele, also called "*Canarium schweinfurthii* Engl." belongs to Buceraceae familly. It is a non conventional oilseed of cultural, economic and nutritional importance in the tropics. *Canarium* tree is found particularly from West to East and South Africa. The fruits are ovoid and their sizes vary from 2 to 4 cm long and from 1.5 to 2 cm wide, depending on the svariety and origin (Kapseu *et al.*, 1996 and Tchiegang *et al.*, 2000). The fruits are composed of a pulpy endocarp and a kernel. They are picked at the mature stage which is indicated by a change of the colour of the epicarp, from green to black. They are consumed after softening in water at 45 °C for 40 min (Tchiegang *et al.*, 2001).

The endocarp tissue, which is the edible part of the fruits, contains a high amount of lipids (36 – 50 % dry weight basis) and carbohydrates (21 – 39 % dry weight basis) in addition to substantial amounts of protein (2.59 – 3.93 % dry weight basis) and dietary fibre (5.73 – 6.40 % dry weight basis) (Tchiegang *et al.*, 2000). The fruits are therefore a complement of the usual base foods as cassava, yam, sweet potatoes, etc... Besides the nutritional aspect, all parts of the plant are used in other to satisfy their needs: when boiled in water, the leaves of the aiele plant constitute a drug against typhoid. The wood is a source of energy. It is highly appreciated in handicraft. The bark is used to treat lung and stomach diseases. The resin extract from the tree, is a gum used to light torch and to treat stomach pains and angina pectoris (*Tchouamo et al.*, 2000).

Although it is known as a multipurpose plant, the major problem with *Canarium* fruits is their preservation after harvesting. Fruits stored at ambient temperature showed a post harvest hardening phenomenon, which reduced their storage durability. The hardening of *C. schweinfurthii* is a serious handicap because the fruit can also become hardened after treatment in the water at 70 °C, thus, making their consumption impossible (Tchiegang *et al.*, 2001). The high perishability of *C. schweinfurthii* is manifested in loss of quality, which is suspected to be induced by the endogenous biochemical reactions of the pulp, leading to textural changes.

To the best of our knowledge, there is no investigation on the biochemical and textural changes of *C. schweinfurthii* fruit during storage. The present study is aimed at providing information concerning the physical and chemical changes associated with the raw, the softened and the hardened pulps of *C. schweinfurthii*, and to determine how they relate to the hardening phenomenon.

Materials and Methods

Materials: Canarium fruits were harvested mature in Bayangam and Babajou (West Cameroon) which are the localities where these fruits are intensively produced. The fruits were then divided into four groups. One group of the fruits was heat treated in water at 45 °C for 40 min, to obtain softened pulps (SF). The second group was heat treated in water at 70 °C for 40 min, to produce hardened pulp (HHF). The third group was stored at room conditions (22 °C) for a period of 7 days to obtain hardened pulp (SHF). The last group was made of raw fruits (RF).

Textural Analysis: The fruits were peeled and the kernel was removed. The sample were then cut into slices of 1 cm diameter, and analysed for their textural properties using an Instron Machine ® texturometer (*Arilest N° 86-104*). The speed was 5mm/mn and the shear force required to cut completely through the slices, as well as the area under the curve were recorded as the level of hardness and adhesiveness respectively on a graphic recorder. Determinations were done in triplicates.

Biochemical Analysis: Samples were analysed for moisture, ash, lipid and protein contents. Total soluble and reducing sugars, starch, lignin and cellulose were also determined. Uronic acids were determined in alcohol insoluble solids (AIS). To obtain AIS, 40 g of pulp were crushed in four volumes of 96 % ethanol, and then filtered through a G_3 sintered glass filter. The insoluble materials were extracted with 64 % ethanol until the filtrate was sugar free. The residue was extracted with methanol/chloroform (1/1, v/v) to remove fats. The insoluble materials were finally air dried over night. Three grams of AIS were treated three times (16, 3 and 3 H) with 120 ml of 0.05 M CDTA (Cyclo hexane diamino tetraacetic acid), pH 4.8 at 4 °C. The combined residue was then extracted sequentially twice (16 and 3 H) at 4 °C with 90 ml of 1 and 4 M KOH containing 0.0126 M $NaBH_4$ each. The CDTA extracts were pooled and extensively dialysed against 0.1 M NaCl, then distilled water and freeze-dried. The KOH extracts were also pooled, brought to pH 5 with pure acetic acid, and extensively dialysed against distilled water, and freeze-dried.

Analytical Methods: The samples were analysed for moisture and ash contents using UICPA methods (UICPA, 1979). Lipids were extracted and estimated by Folch method, using chloroform / methanol (2/1, v/v) (Omotia, Okyi, 1987). Starch was determined using enzymatic method (á-amylase from *Aspergillus oryzae*, Sigma) (AACC, 1990). Proteins were determined by Kjeldahl method (*GERHARDT*, Vapodest 4S, Paris France). Total alcohol-soluble and reducing sugars, lignin and cellulose were determined by spectrophotometric procedure using an Ultra-Spec 4000 spectrophotometer (Pharmacia Biotech) (Dubois *et al*., 1956; Updegraff, 1969 and Morrison, 1972). Uronic acids were determined by m- hydroxydiphenyl method after acid hydrolysis (Blumenkrants and Asboe-Hansen, 1973).

Statistical Analysis: The data obtained from the physicochemical analyses were statistically analysed using Statgraphics (Graphics Software system, USA). Comparison between sample treatments and the results were done using analysis of variance (ANOVA) with a probability of 0.05.

Results and discussion

Textural Studies: The Instron Machine ® Texture Analyser was used for the measurement of the textural properties of *C. schweinfurthii* pulp. Measurements were made on raw freshly harvested fruits (RF), on hardened fruits after 7 days of storage (SHF), on softened fruits (SF, heat treated at 45 °C for 40 min) and on heat hardened fruits (heat treated at 70 °C for 40 min), to find the changes occurring in the textural properties of pulp. Shear forces generated by the Texture Analyser, were recorded as the hardness of the samples and the area under the curves was recorded as adhesiveness.

Hardness of the pulp of *C. schweinfurthii* fruits increased significantly during storage (Fig. 1), while adhesiveness increased during heat treatment (Fig. 2). Heat treated hardened fruits showed variation in hardness. As compared to raw fruits (15.83 N) and stored hardened fruits (20.71 N), heat hardened fruits showed a comparatively lower level of hardness (5.41 N). This level of hardness of heat treated hardened fruits, is higher than the hardness of softened fruits (0.77 N), thus, making the consumption of heat hardened fruits impossible. Consistent increases in shear force (hardness) during storage resulting from the hardening of *C. schweinfurthii* fruits after harvest, could be attributable to increase in cell wall stiffness and subsequent strengthening of cell wall bonding during storage. Low temperature treatment (45 °C) brings about softening of the pulp (Tchiegang *et al*., 2001) and adhesiveness also increased in the softened fruits. Low temperature of treatment (45 °C) can then be used to delay the hardening after harvesting. The hardness of heat treated (70 °C) fruits is probably due to the increase in temperature, which might have been facilitated by biochemical reactions leading to the hardening phenomenon in fruits.

Physicochemical Analyses: The moisture content of *C. schweinfurthii* pulp showed no significant difference (Table I) between raw fruits and heat treated fruits (softened fruits and heat hardened fruits). Differences observed were on stored hardened fruits. The moisture level of the fruits kept at 22 °C decreased from 49.32 to 39.90 % after 7 days of storage. Within 7 days of storage at 22 °C, about 19 % moisture was lost in the fruits. The fruits undergo rapid dehydration after harvesting, leading to hardening phenomenon. The effect of this dehydration might have brought the cell wall polysaccharides closer together, permitting greater interaction, resulting to increase cell rigidity during storage of fruits.

The crude protein contents of *C. schweinfurthii* fruits were 7.56, 7.97, 8.05 and 8.38 % (Table I) of dry weight for Heat Hardened fruits, Raw fruits, Stored Hardened fruits and Softened fruits respectively. Analysis of variance showed that the treatment condition of the fruits (heat treated or stored for 7 days) does not affect significantly their protein contents.

C. schweinfurthii generally has considerably high fat content. The mean fat contents were found to be 37.50 – 43.96% respectively for Stored hardened fruits and softened fruits (Table I). Fat contents of fruits were affected by storage and

Table 1: Chemical composition of C. *schweinfurthii* pulp (g/100g dw)

	RF	SF	HHF	SHF
Moisture (%) a	49.32±0.92 ^{b}	48.41±0.73 ^{b}	50.49±0.77 ^{b}	39.90±0.63
Ash (%) a	3.89±0.31 ^{a}	3.00±0.82 ^{a}	3.31±0.27 ^{a}	3.55±0.20
Protein (%) a	7.97±0.38 ^{a}	8.38±0.42 ^{a}	7.56±0.78 ^{a}	8.05±0.25
Fat (%) a	38.00±0.96 ^{a}	43.96±1.08 ^{b}	42.50±0.56 ^{b}	37.50±0.85
Minerals				
Na (%) a	0.03±0.01 ^{a}	0.04±0.01 ^{a}	0.05±0.01 ^{a}	0.03±0.01
K (%) a	0.55±0.02 ^{a}	0.54±0.01 ^{a}	0.56±0.02 ^{a}	0.57±0.02
Ca (%) a	0.29±0.01 ^{b}	0.30±0.01 ^{b}	0.30±0.01 ^{b}	0.20±0.01
Mg (%) a	0.13±0.01 ^{ab}	0.15±0.01 ^{b}	0.13±0.01 ^{ab}	0.10±0.01

Values within each row with the same letter are not significantly different ($P<0.05$)
Values within each row with different superscripts are significantly ($P<0.05$) different
RF= Raw Fruit; SF = Softened Fruit; HHF = Heat Hardened Fruits; SHF = Stored Harden Fruit

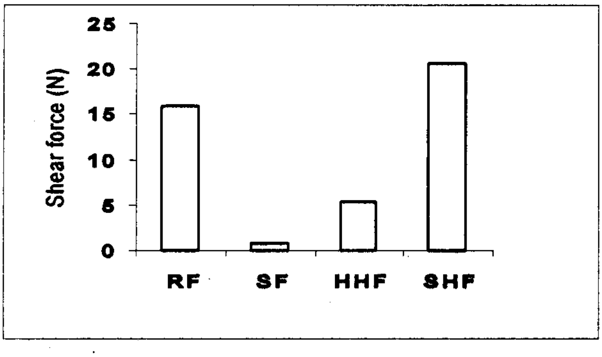

Fig. 1: Changes in shear force (hardness) of *C. schweinfurtii* Eng. fruits during heat treatment and storage
RF= Raw Fruit; SF = Softened Fruit; HHF = Heat Hardened Fruit; SHF = Stored Hardened Fruit.

temperature of treatment. There was a decrease in Fat level after 7 days of storage. Heat treated fruits (45 or 70 °C) yielded more oil (42.50 – 43.96 % DW) than raw fruits (38 %), and these results mean that the extraction yield is significantly influenced ($P<0.05$) by heat treatment.

The mean ash contents for fruits studied ranged from 3 to 3.89 g/100g (Table I). Analysis of variance showed that the treatment conditions of the fruits (heat treated or stored for 7 days) does not affect significantly ($P<0.05$) their ash contents. Similarly, mineral contents (Na, K, Ca, Mg) of the fruits studied (Table 1) were nod affected by heat treated condition.

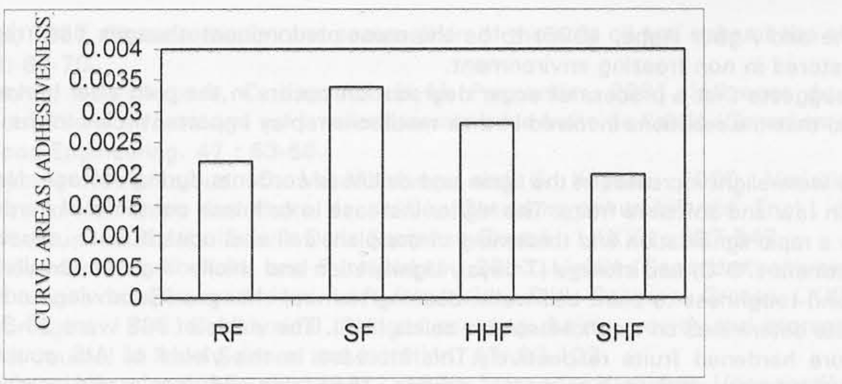

Fig. 2: Changes in curve areas (adhesivity) of *C. schweinfurtii* Eng. fruits during heat treatment and storage
RF= Raw Fruit; SF = Softened Fruit; HHF = Heat Hardened Fruit; SHF = Stored Hardened Fruit

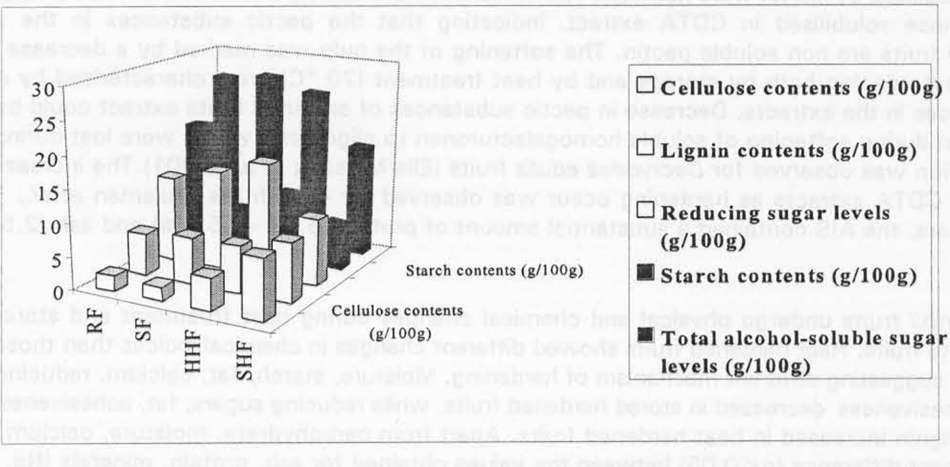

Fig. 3: Changes in polysaccharides contents of *C. schweinfurtii* Eng. fruits during heat treatment and storage
RF= Raw Fruit; SF = Softened Fruit; HHF = Heat Hardened Fruit; SHF = Stored Hardened Fruit

Changes in starch contents of fruits were affected by storage and heat treatment.There was a general decrease in starch levels of all the samples (Fig. 3). The rates of decrease were higher in the heat softened fruits and stored hardened fruits than in heat hardened fruits.

Table 2: Uronic acid contents of fractions extracted from cell wall materials of *C. schweinfurthii* fruits

	Extracts		
	0.05 M CDTA	1 M KOH	4M KOH
Uronic acid (% of the fraction)			
RF	0.74 ± 0.01^{b}	5.23 ± 0.09^{b}	0.71 ± 0.00^{a}
SF	0.52 ± 0.02^{a}	2.59 ± 0.04^{a}	1.09 ± 0.01^{d}
HHF	0.70 ± 0.04^{b}	2.59 ± 0.08^{a}	0.90 ± 0.03^{b}
SHF	0.91 ± 0.05^{c}	2.59 ± 0.01^{a}	1.01 ± 0.01^{c}

Values within each colon with the same letter are not significantly different ($P<0.05$)
Values within each colon with different superscripts are significantly ($P<0.05$) different
HHF = Heat Hardened Fruit; RF= Raw Fruit; SF = Softened Fruit; SHF = Stored Harden Fruit

The pulp of *C.schweinfurthii* fruits showed a higher concentration of total soluble sugars, with mean values of 26.17 % of DW for raw fruits. The mean reducing sugar contents of *C. schweinfurthii* were found to be 12.96 % of DW for raw fruits. Changes in sugar contents were observed for the samples after treatment. In general, the total soluble sugars and reducing sugars decreased after storage, while the reducing sugar levels increased in heat hardened fruits (Fig. 3). The increasing levels of reducing sugars in the heat treated fruits are suspected to be brought about by the breakdown and subsequent hydrolysis of starches in to sugars. Similar trends have been reported (Treche and Agbor Agbe, 1996) to be the most predominant changes occurring in yam tubers after harvest, when stored in non freezing environment.

This however suggests that a process of sugar degradation occurs in the pulp after harvest and during storage, and it is believed that the reactions involved in their metabolism play important roles in the quality changes of the fruits.

Generally, there were slight increases in the lignin and cellulose contents during storage. No significant difference was observed on raw and softened fruits. The higher increase in cellulose contents of hardened fruits might have been caused by a rapid lignification and thickening of the plant cell wall constituents, causing rigidity of the fruits during heat treatment (70°C) and storage (7 days). Lignification and thickening of cellwalls have been reported to confer rigidity and toughness to plant cell walls, causing textural changes (Goodwing and Mercer, 1992).

Uronic acids were determined on alcohol-insoluble solids (AIS). The yields of AIS were 28-35 % of the dry weight of raw and store hardened fruits respectively.This increase in the yields of AIS could be attributed to the dehydration of the fruits which occurs during storage. The yields and uronic acid percentages of extracts are presented in Table II, for raw, softened and hardened fruits.

Uronic acid contents in CDTA extracts account for 0.52 to 0.91% of AIS. For all the samples, the amounts of uronic acid solubilised by CDTA were not high. The amounts of uronic acid solubilised in alkali extract were greatly superior to those solubilised in CDTA extract, indicating that the pectic substances in the cellwall of *C. schweinfurthii* fruits are non soluble pectin. The softening of the pulp was marked by a decrease in uronic acid content, while hardening both by storage and by heat treatment (70 °C) were characterised by a high level of pectic substances in the extracts. Decrease in pectic substances of softened fruits extract could be attributed to the degradation during softening of soluble homogalacturonan to oligomers, which were lost during dialisis step. Simular evolution was observed for *Dacryodes edulis* fruits (Ella Missang *et al.*, 2001).The increasing content of uronic acid in CDTA extracts as hardening occur was observed for olive fruits (Huisman *et al.,* 1996). Beside cellwall polymers, the AIS contained a substantial amount of protein (6.88 –8.53 %) and ash (2.50 –5.29 %).

Conclusion

C. schweinfurthii fruits undergo physical and chemical changes during heat treatment and storage, leading to hardening of the fruits. Heat hardened fruits showed different changes in chemical indices than those stored at 22 °C for 7 days, suggesting different mechanism of hardening. Moisture, starch, fat, calcium, reducing sugars, total sugars and adhesiveness decreased in stored hardened fruits, while reducing sugars, fat, adhesiveness, uronic acid, cellulose and lignin increased in heat hardened fruits. Apart from carbohydrate, moisture, calcium and fat, there was no significant difference ($p<0.05$) between the values obtained for ash, protein, minerals (Na, K, Mg) of the samples. The hardening of fruits after storage or heat treatment (70 °C) is brought about by a series of biochemical reactions occurring in fruits. Low temperature of treatment (45 °C) involves softened pulp and adhesiveness also increased in softening fruits. Low temperature of treatment (45 °C) can then be used to delay the hardening process after harvesting.

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