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Effect of tuber storage and pre- and post-blanching treatments on the physicochemical and pasting properties of dry yam flour

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Abstract

In West Africa, yams are often processed into dry flour that is consumed as a thick paste (amala), the main quality attributes of which are colour, texture and taste. The traditional process consists of peeling, blanching and drying the fresh tubers. The fresh tubers may also be stored for several months before processing. This study was undertaken to test whether fresh tuber storage and pre- and post-blanching treatments (1-h storage of sliced fresh yam pieces in air or water and steeping in the blanching water, respectively) affect the physicochemical characteristics and pasting behaviour of dry yam flour produced from D. rotundata cultivars. Both pre-blanching treatments significantly reduced only peroxidase activity, while the post-blanching treatment significantly reduced polyphenoloxidase activity. Despite the effects on enzymatic activities, neither pre- nor post-blanching treatments had any significant effect on the amala brown index or the flour total phenol content. However, peak areas of three major cinnamic acid compounds, detected by HPLC, decreased continuously with steeping time after blanching, while fresh tuber storage led to a dramatic reduction in amala paste viscosity.

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1. Introduction

Yam (Dioscorea spp) is one of the major staple foods in the Gulf of Guinea countries. To overcome the high perishability of fresh yam, a large proportion is processed into dry flour, mainly in Nigeria and Benin [\(Bri](#page-8-0)[cas et al., 1997; Onayemi & Potter, 1974](#page-8-0)). Dried yam is less perishable and can be consumed throughout the year, mainly as a thick paste called amala (Akingbala, Oguntimein, & Sobande, 1995; Akissoe, Hounhouigan, Bricas, Vernier, Nago, & Olorunda, 2001). The main quality attributes of amala are colour, texture and taste. Most consumers prefer a brownish, elastic, non-sticky amala with a slightly sweet taste, while a slightly bitter taste is also tolerated ([Akissoe et al., 2001; Hounhoui](#page-8-0)[gan, Kayode´, Bricas, & Nago, 2003; Mestres, Dorthe,](#page-8-0) [Akissoe, & Hounhouigan, 2003\)](#page-8-0).

Traditionally, tubers are processed at farm level by peeling, sometimes followed by slicing, blanching in hot water (at around $63-65$ °C for 15–50 min) and sun-drying. Recent studies [\(Akissoe et al., 2001](#page-8-0)) have shown that yam cultivar characteristics and blanching and drying operations directly influence amala quality. Amala colour appears to be linked, in particular, to the total phenol content of the flour and to the peroxidase activity of the fresh yam tubers [\(Akissoe, Hounhouiyon,](#page-8-0) [Mestres, & Nago, 2003; Izundu, 1995\)](#page-8-0). In addition, some variations occur in the dry yam process. Because of the time required to peel tubers by hand, they are often kept in water or left on the ground before blanching. This may affect the colour of the yam as polyphenoloxidase (PPO) and peroxidase (POD) are active during this step ([Ikediobi, Chelvarajan, & Agwu,](#page-8-0) [1989\)](#page-8-0). Preliminary research also indicated that yam tubers are sometimes stored overnight in the blanching bath before drying. PPO inactivation and the starch annealing process that occur during blanching may therefore continue during steeping, thus affecting amala quality ([Akissoe et al., 2003](#page-8-0)).

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Furthermore, because of the volume of farm work during yam harvesting (which coincides with the harvesting of sorghum and cotton and with land cleaning), tubers are often stored on the farm for some weeks before being dried. Martin and Ruberté (1975) and [Onayemi and Idowu \(1988\)](#page-8-0) reported that the levels of polyphenolic and glycoalkaloid substances in stored tubers increased and became concentrated in the head regions, to which they attributed the tendency of the latter to discolouration. [Asemota, Wellington, Odutuga,](#page-8-0) [and Ahmad \(1992](#page-8-0)) observed that peroxidase and odiphenolase activity increased during the third week of storage. [Izundu \(1995\)](#page-8-0) also noted that yam flour, prepared soon after harvesting, did not display any significant browning because of negligible peroxidase activity. In contrast, polyphenoloxidase activity and starch content decreased with long-term storage and polyphenoloxidase activity was low in old tubers [\(Adamson & Abigor, 1980; Ikediobi & Oti, 1983;](#page-7-0) [Ravindran & Wanasundera, 1992\)](#page-7-0).

The present study was undertaken to determine the changes that occur when peeled yam tubers are stored in air or water before blanching and/or are left to steep in the blanching bath, and to evaluate the effect of storage for a varying number of weeks on the colour and pasting properties of dry yam flour and paste.

2. Materials and methods

2.1. Plant material

Experiments were conducted in 2001 and 2002 using two Dioscorea rotundata cultivars (Deba and Banioure). The tubers came from two different sources: the Deba tubers used for the storage experiment were obtained from the experimental station of the International Institute of Tropical Agriculture (IITA) in Southern Benin in December 2000. The other experiments were performed in 2002 using Deba and Banioure tubers purchased from farmers in Northern Benin in December 2001. Only tubers with no sign of damage or insect attack were selected for the experiments. Just after harvesting, the fresh tubers were cleaned of dirt and packed in a well-ventilated room on a platform 1 m above the floor. The temperature range and the relative humidity range of the storage room were $26-29$ °C and $55-80\%$, respectively (Hygrothermograph, Model NSII-Q, Sato Keiryoki MFG Co. Ltd., Japan).

2.2. Experimental design

2.2.1. Effect of fresh tuber storage

A randomly-selected batch of Deba cv tubers (about 5–6) harvested in 2000 was removed from the storage room after 7, 21, 35, 49, 63 and 91 days of storage. The tubers began sprouting after 70 days and the emerging sprouts were not broken off. Immediately after removal, each batch was peeled under water (25–27 \degree C) and cut into 30 mm thick slices, using a cylinder cutting box. About one third of the slices were freeze-dried as a control and the other two-thirds were blanched using a thermostatically-controlled water bath set at 65 \degree C for 20 min. The blanched slices were then equilibrated in water at $28-30$ °C for 5 min and freeze-dried. Each batch was then crushed in a mortar and milled in a laboratory centrifuge mill (Retsch Haan, Germany) fitted with a 0.2 mm screen. The flour obtained from each batch was then packed in a polyethylene bag and stored at 4° C until analysis.

2.2.2. Effect of pre- and post-blanching treatments

Deba and Banioure cv tubers, harvested in 2001, were used for these experiments, which were performed 15 days after harvesting. For the experiment to determine the effect of storing peeled tubers before blanching, 3–4 tubers of each cultivar were peeled and sliced as described above. Each batch was left for one hour either in water or in the open air at ambient temperature (28– 30° C), before being processed as above.

The experiment to determine the effect of steeping in the blanching bath was performed on 5-6 tubers of each cultivar, which were peeled and sliced as previously. The slices were then blanched at $65 °C$ for 20 min and a control sample was removed and immediately freezedried. The remainder were steeped in the blanching bath (with the heater switched off) and samples were removed after 2 h, 4 h, 6 h, and 12 h. Each sample was immediately freeze-dried.

2.3. Methods

2.3.1. Enzymatic activity and total phenol determination

Polyphenoloxidase (PPO), peroxidase (POD), and total phenol (PT) content were determined using the methods described by [Mestres et al. \(2003\)](#page-8-0), measuring the oxygen consumption and discoloration kinetics at 460 nm with catechol as substrate and optical density at 760 nm after reaction with Folin reagent.

2.3.2. Determination of phenolic compounds by HPLC

HPLC analyses were carried out on an Altima C18 5U column (250 mm \times 4.6 mm). The solvent system used was derived from that of [Guyot, Marnet, Laraba,](#page-8-0) [Sanoner, and Drilleau \(1998\)](#page-8-0) and consisted of two solvents, solvent A (acetonitrile/acetic acid/water; 3/2.5/ 95.5) and solvent B (acetonitrile/acetic acid/water; 95.5/ 2.5/3). The gradient applied was: 0 min, 100% A, isocratic; 0–40 min, 70% A, linear; 40–43 min, 0% A, linear; 43–45 min, 0% A, isocratic; 45–48 min, 100% A, isocratic; 48–55 min, 100% A, isocratic. The detection was performed at 280 and 320 nm and recorded using Kroma System 2000 software (Bio-Tek Instruments). For sample preparation, 250 mg of yam flour were vigorously suspended in 1 ml methanol/HCl (1.5N) (85/15, v/v) and stirred at ambient temperature (27–30 °C) for 30 min. The suspension was then centrifuged at 7000 g for 5 min. The supernatant was filtered through $0.45 \mu m$ pore size filters and $100 \mu l$ were then injected into the HPLC system.

2.3.3. Hunter Lab colour coordinates

The colours of the pastes (obtained with a Rapid Visco Analyser as described below) were measured using a Minolta CR-210 portable chromameter (illuminant D65 CIE 1976). The Hunter Lab colour coordinates system L^* , a^* and b^* values were recorded and the brown index was calculated as (100-L*).

2.3.4. Pasting behaviour

Pasting properties were determined using a Rapid Visco Analyser (RVA, Newport Scientific, Narrabeen, Australia) on an 8% dry matter suspension. The suspension was heated from 35 °C to 95 °C at a rate of 6 °C min⁻¹, maintained at 95 \degree C for 4 min, then cooled to 50° C at the same rate. Peak viscosity, viscosity at the start of the 95 °C plateau (V95b), viscosity at the end of the 95 \degree C plateau (V95e) and end viscosity, after cooling to 50 \degree C(V50), were measured.

2.3.5. Swelling power and solubility

The swelling power and solubility procedures described by Mestres, Nago, Akissoë, and Matencio (1997) were modified, with a dry matter concentration of 4% (wb; 1.2 g of dry matter dispersed in distilled water to give a total mass of 28 g). The suspension was heated from 35 °C to 95 °C at 6 °C min⁻¹ and held at 95 °C for 1 min using a RVA. The heated suspension was then centrifuged at 3,000 g for 15 min at ambient temperature.

2.4. Statistical analysis

Analysis of variance and correlation and regression analyses were performed using Statitcf software (ITCF, Boigneville, France).

3. Results

3.1. Colour parameters and related compounds

Fresh freeze-dried Deba gave a higher amala brown index than Banioure (46.5 versus 41.0). It presented both a concomitantly higher total phenol content and concomitantly higher PPO and POD activities: 1.41 µmole gallic acid equivalents g^{-1} , 25.3 µMol [O₂] min^{-1} g⁻¹ and 85 mDO min⁻¹ g⁻¹ versus 0.85, 17.9 and 61 for Deba and Banioure, respectively. The brown index and total phenol content were not significantly affected, either by pre-treatment (slices exposed to air or left in water for one hour) or by blanching at 65° C for 20 min (Table 1) for both cultivars. PPO activity was not significantly affected by either method of pre-treatment but decreased by 60% after blanching. POD activity was significantly reduced by pre-treatment, whatever the method used (air or water), and also by blanching.

Steeping after blanching had no significant effect on either the amala brown index or the flour total phenol content [\(Table 2](#page-3-0)). However, the total phenol content of the steeping water increased slightly over the first four hours and decreased thereafter: 0.9 µmol gallic acid equivalents ml⁻¹ just after blanching, 1.7 μ mol ml⁻¹ after 4 h of steeping and 0.7 μ mol ml⁻¹ after 12 h of steeping. PPO activity decreased significantly during steeping, particularly over the first 2 h. POD activity was very low just after blanching and ceased completely after 2 h.

Table $1 \cdot$

Values in the same column with different letters are significantly different $(P < 5\%)$.

^a Standard deviation of the residual.

Two groups of polyphenolic compounds were seen on HPLC chromatograms of yam extracts, based on their maximum absorption wavelengths (λmax) : flavanols with a λ max of around 280 nm and cinnamic acid compounds with a λ max of around 320 nm. Only the results for cinnamic acid compounds are presented. Four major peaks were observed for Deba extracts with retention times of 17.1, 25.9, 27.6 and 32.2 min (Fig. 1). The last three peaks decreased continuously with steeping time [\(Fig. 2\)](#page-4-0).

The amala brown index increased from 50 to 52 when fresh tubers were stored for 3 weeks. It then decreased steadily to 47 after 13 weeks. No clear trend was observed in the evolution of the total phenol content during fresh tuber storage, whereas PPO and POD activities [\(Fig. 3\)](#page-4-0) showed a plateau between the fifth and ninth weeks of storage, then decreased when sprouting began (after 10 weeks).

3.2. Pasting behaviour

Fresh freeze-dried Deba gave a paste with higher viscosities and a higher swelling power and solubility

Table 2

Effect of steeping time after blanching on some physico-chemical characteristics of yam flours (mean values for Deba and Banioure cultivars)

Steeping duration (H)	Brown index $(100-L)$	Total phenols μ mols g ⁻¹	PPO activity [O2] μ mols min-1 g-1	POD activity mDO min ⁻¹ g^{-1}
Ω	43.0	1.12	10.2a	4.6
	41.2	1.05	8.5 ab	
4	41.5	0.98	8.7 ab	
6	41.9	1.08	8.4 ab	
12	40.3	0.90	8.0 _b	
SDR ^a	1.3	0.1	0.5	1.6

Values in the same column with different letter are significantly different $(P < 5\%)$.

^a Standard deviation of the residual.

Fig. 1. Chromatograms for Deba flour extracts after blanching (upper curve) and after 12 h of steeping (lower curve).

index than fresh freeze-dried Banioure: V50 and swelling power were 249 RVU and 20.0 g/g (db) for Deba, versus 221 RVU and 15.3 g/g (db) for *Banioure*. A significant pre-treatment effect was observed only on V50 for unblanched samples: yam exposed to air for 1 hour had a higher V50 value [\(Table 3\)](#page-5-0). In addition, blanching significantly reduced the V95b and V50 values of the air-exposed samples. No significant difference was found between treatments as far as swelling power and solubility index were concerned.

There was no significant effect on paste viscosities at the 5% level of steeping time. However, V95b decreased with steeping time (from 180 to 149 RVU within 12 h), the effect being significant only at the 7% level [\(Table 4\)](#page-5-0).

Fig. 2. Effect of steeping time after blanching on peak areas measured at 320 nm by HPLC for Deba flour extracts.

Fig. 3. Evolution with storage of PPO and POD activities of fresh Deba tubers.

Swelling power decreased from 15.9 g g^{-1} to 14.2 g g^{-1} after 2 h of steeping then remained virtually constant. In addition, it was observed that the steeping water became turbid and creamy as a result of matter loss. Paste viscosity parameters (V50, Vpeak, V95b) decreased with length of tuber storage, as shown in Fig. 4 for V50.

4. Discussion

Fresh tuber colour and related compound characteristics of Deba and Banioure were consistent with those previously observed [\(Akissoe et al., 2003\)](#page-8-0), the former having higher PPO activity and giving amala with a higher brown index. However, the classification of the

V50 : viscosity at 50 °C during cooling (RVU); Values of any viscosity parameter with different letter are significantly differents ($P < 5\%$); Vpeak : peak viscosity (RVU).

^a Standard deviation of the residual.

Table 4

Effect of steeping time after blanching on pasting behaviour of yam flours (mean values for Deba and Banioure cultivars)

Values in the same column with different letter are significantly different $(P < 5\%)$.

^a Standard deviation of the residual.

Fig. 4. Effect of storage of fresh Deba tubers on amala paste viscosity (V50).

two cultivars was reversed as far as phenol content and POD activity were concerned. This may be due to intracultivar variability linked to agro-ecological conditions. [Osagie and Opoku \(1984\)](#page-8-0) reported considerable variability in PPO activity, total phenol and intensity of browning in Dioscorea species cultivars, but were unable to identify the causative factors.

Pre-treatment affected only POD activity, reducing it by 60% after 1 h, either in water or in air, whereas PPO activity remained unchanged over the same time. In contrast, [Ikediobi et al., 1989](#page-8-0) observed a slight increase in PPO activity and a dramatic increase in POD activity after tuber wounding. However, these increases were noticeable only after 3–4 days. In our case, peeling and cutting can be considered as wounding but the length of the experiment (one hour) was certainly not sufficient to promote PPO and POD synthesis.

POD activity ceased almost completely after blanching, as already observed by [Akissoe et al. \(2003\),](#page-8-0) so subsequent steeping had no effect on this characteristic. PPO activity decreased after blanching but did not disappear completely, indicating its relative stability to heat, in contrast to POD. Osagie and Opoku (1984), in fact, reported a residual PPO activity of about 20% after incubation at 70 \degree C for 35 min. PPO activity continued to decrease during steeping, particularly during the first 2 h after heating. This suggests that blanching was not complete after 20 min at 65 \degree C and could be

Fig. 5. Relation between amala brown index and total phenol content of flour.

Fig. 6. Relation between apparent viscosities (V95b, \Diamond ; V50, \Box) and swelling power of yam flours.

improved by steeping. However, higher blanching temperatures would appear to be necessary to inactivate PPO completely: [Ikediobi and Obasuyi \(1982\);](#page-8-0) [Ozo and](#page-8-0) [Caygill \(1985\)](#page-8-0) observed that PPO activity of purified yam disappeared only above 80 °C.

The plateau of PPO and POD activities between the fifth and the ninth weeks of storage is in global agreement with the results of [Asemota et al. \(1992\)](#page-8-0) and [Ike](#page-8-0)[diobi and Oti \(1983\),](#page-8-0) who observed an increase in both enzyme activities during the first three weeks of storage, then a decrease after the fifth week, for D. rotundata, cayenensis and alata tubers. In the case of D. dumetorum, however, [Izundu \(1995\)](#page-8-0) reported a continuous increase in peroxidase activity up to the twenty-fifth week of storage. In agreement with [Ikediobi and Oti](#page-8-0) [\(1983\)](#page-8-0), PPO activity was lower in old tubers than in freshly harvested ones, as a result of a fall in PPO activity after the start of sprouting ([Fig. 3](#page-4-0)). [Ikediobi and](#page-8-0) [Oti \(1983](#page-8-0)) accordingly inferred that the sprouting mechanism was associated with a reduction in PPO activity.

Unlike enzyme activities, the flour total phenol content and amala brown index that may result, at least in part, from such activities [\(Izundu, 1995; Akissoe et al.,](#page-8-0) [2003\)](#page-8-0) are not significantly affected by any treatment. This may be explained by considering two adverse phenomena: the first is an increase in phenol content due to enzyme or chemical oxidation and the second is their solubilization in the steeping water, which was measured. As has already been observed [\(Akissoe et al.,](#page-8-0) [2003; Mestres et al., 2003; Osagie & Opoku, 1984](#page-8-0)), the brown index of amala is closely correlated $(r=0.88)$ with the phenol content of the flour ([Fig. 5\)](#page-6-0). This confirms the high contribution of polyphenols to the browning of the amala paste.

The detection of flavanol compounds by HPLC in Deba and Banioure extracts is in agreement with [Ozo et](#page-8-0) [al. \(1984\),](#page-8-0) who identified catechin in extracts of various Dioscorea species. We also detected cinnamic compounds, four of which could be quantified. By comparison, [Martin and Ruberte \(1976\)](#page-8-0) showed only one cinnamic compound among the thirteen phenolic compounds extracted from D. alata. The decrease in cinnamic compounds during steeping may be due to their solubilization in the steeping water. It can also be hypothesized that part of the cinnamic compounds may be involved in a polymerization process during steeping, thus explaining their decrease, whereas the total phenol content remains constant.

The pasting behaviour of the fresh Deba and Banioure tubers was very different from that observed by [Akissoe](#page-8-0) [et al. \(2003\):](#page-8-0) V50 and swelling power mean values were higher in the present study (235 RVU and 17.7 g g^{-1} , respectively, versus 157 RVU and 13.3 $g g^{-1}$), whereas solubility was lower (8.6 versus 12.1 mg ml⁻¹). This could be linked to the agro-ecological conditions in

which the tubers were grown: both cultivars used to measure pasting behaviour in the present study were in fact collected from farmers in Northern Benin, whereas, for the previous study, they had been obtained from an experimental station in Southern Benin. The end viscosity values measured during the storage experiment [\(Fig. 4](#page-5-0)) appear to support this hypothesis, as they were close to those obtained in the previous study and, in this case, the Deba tubers came from the same experimental station.

A slight decrease in pasting viscosity (particularly V95b) was observed after blanching and steeping. Swelling power showed a similar pattern. These observations suggest that an annealing process occurs during both unit operations, as already indicated by the increase in the gelatinization temperature after steeping in a previous experiment [\(Akissoe et al., 2003\)](#page-8-0). As already reported in this previous study, there was a highly positive significant correlation between paste viscosity and swelling power: the correlation coefficients were 0.92 and 0.76 for V95b and V50, respectively [\(Fig. 6\)](#page-6-0). It should be noted that the viscosity calculated according to the formula of [Akissoe et al. \(2003\)](#page-8-0), using the swelling power and solubility data, was close to the observed values: e.g., for fresh freeze-dried tubers, the calculated V50 was 246 RVU (swelling power: 17.7, solubility index: 8.6) versus an observed mean value of 235 RVU. This indicates that the higher viscosities observed for yam obtained from Northern Benin were linked to intrinsic properties of starch which depend on agroecological conditions. It also suggests that swelling power, and to a lesser extent solubility, is sufficient to describe the pasting behaviour of yam flour.

Pasting viscosity fell during tuber storage ([Fig. 4\)](#page-5-0). The use of stored yam tubers may therefore result in an amala with a lower-quality texture. The fall in pasting viscosity may be linked to a decrease in starch content due to hydrolysis, as has already been observed [\(Ikediobi & Oti, 1983; Onayemi & Idowu, 1988\)](#page-8-0). It may also be due to the annealing process.

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