



Physicochemical and functional properties of starches of two quality protein maize (QPM) grown in Côte d'Ivoire

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ABSTRACT

Objective: The aim of this study was to investigate physico-chemical and functional properties of starches from two novel (white and yellow QPM) varieties of maize cultivated in Côte d'Ivoire in order to explore their potential use in food and non food industries.

Methodology and results: Starches from four varieties of maize, namely two ordinary (white and yellow varieties) and two QPM (white and yellow varieties) were extracted and their physico-chemical and functional characteristics were determined. Statistical analyses were performed on data obtained. Protein and fat contents ($0.35-0.37 \pm 0.06\%$; $0.53-0.57 \pm 0.05\%$) of white and yellow QPM starches were lower than that ($0.38-0.39 \pm 0.06\%$; $0.63-0.66 \pm 0.05\%$) of white and yellow ordinary maize. The particle size of the four starches varied from 4.75 to 22.79 μm for white QPM; 5.28 to 22.62 μm for white ordinary maize; 5.98 to 22.88 μm for yellow QPM and 5.63 to 21.47 μm for yellow ordinary maize. Solubility white QPM starch showed the highest solubility (19.43%) and yellow QPM was characterized by highest value (20.39g/g) of swelling power. The percentage of water expelled from white QPM starch was 40.39% at -15°C and 37.05% at 4°C after the first week of storage while yellow QPM starch was characterized by syneresis values of 40.09% at -15°C and 36.92% at 4°C . The paste clarity of all starches decreased quickly during the first week and then stabilized at about 0.74 to 1%.

Conclusions and application of findings: Starches from white and yellow QPM present better properties than ordinary maize and could be a valuable alternative for exploitation in food and non-food industries.

Key words: QPM maize, starch, physicochemical characteristics, morphology, retrogradation.

INTRODUCTION

Maize (*Zea mays* L.) is the third most important cereal crop and major source of energy (starch) and other nutrients for human and livestock in the world (Jompuk *et al.*, 2011). Maize grain accounts for about 15 to 56% of the total daily calories in diets of people in about 25 developing countries, particularly in Africa and Latin America (Prasanna *et al.*, 2001). With regard to biochemical composition, ordinary or

classic maize has low protein quality. Indeed, maize proteins have poor nutritional value for humans because of reduced content of essential amino acids such as lysine which average (about 2%) is less than one-half of the concentration recommended for human nutrition (FAO, 1992). To improve the poor nutritional protein value of maize grains, quality protein maize (QPM) has been developed by

combining the genetic systems of the gene mutant *opaque-2* ($\delta 2$) and genetic endosperm modifiers (Bello *et al.*, 2012). Products obtained from processing of both ordinary and quality protein maize include starch, high-fructose corn syrup and livestock feed (Kolawole & Titilayo, 2012).

Corn starch is a valuable ingredient to the food industry, being widely used as a thickener, gelling agent, bulking agent and water retention agent (Singh *et al.*, 2003). Normal starch consists of about 75% branched amylopectin and about 25% amylose, that is linear or slightly branched. Amylose is a linear polymer composed of glucopyranose units linked through α -D-(1-4) glycosidic linkages, while the amylopectin is a branched polymer with one of the highest molecular weights known among naturally occurring polymers (Karim *et al.*, 2000). Starch granules swell when heated in excess water and their volume fraction and morphology play important roles in the rheological behaviour of starch dispersions (Da Silva *et al.*, 1997). Starch retrogradation has been defined as the process, which occurs when the molecular chains in gelatinized starches begin to reassociate in an ordered structure (Sandhu & Singh, 2007). During retrogradation, amylose forms double-helical associations of 40–70 glucose units whereas amylopectin crystallization occurs by re-association of the outermost short branches (Ring *et al.*, 1987). Although both amylose and amylopectin are capable

of retrograding, the amylopectin component appears to be more responsible for long-term quality changes in foods (Miles *et al.*, 1985). Starch paste behaviour in aqueous systems depend on the physical and chemical characteristics of the starch granules, such as mean granule size, granule size distribution, amylose/amylopectin ratio and mineral content (Madsen & Christensen, 1996). Several workers have characterized the pasting properties of starches from different corn types and observed considerable variability in these properties (Seetharaman *et al.*, 2001; Ji *et al.*, 2003). In addition, the effect of corn types on the physicochemical, thermal, morphological and rheological properties of corn starches has been studied by Sandhu *et al.* (2004). This study has revealed variations on functional properties of corn starches depending on the genotype, biological origin and cultural practices. However, if several studies have been carried out on various kinds of starches (Amani *et al.*, 1993; Dadié *et al.*, 1998; Gnakri, 1993; Sidibé *et al.*, 2007), there are no scientific data to the best of our knowledge, on the physicochemical characteristics of starch isolated from classic and quality protein maize grown in Côte d'Ivoire. Therefore, the aim of this work is to investigate the physicochemical and functional properties of these starches in order to explore and discuss their potential use in food and non-food industries.

MATERIALS AND METHODS

Raw material: Four varieties of maize, namely two ordinary (white and yellow varieties) and two QPM (white and yellow varieties) were obtained from the collection of the National Rice Program (NRP) of Yamoussoukro city (Côte d'Ivoire).

Starch isolation: Starches were extracted according to Delpuech *et al.* (1978) with few modifications. Samples of 500 g of maize kernels were rinsed thoroughly with distilled water and steeped in sodium metabisulphite solution (0.1%) overnight in refrigerator. The endosperms obtained were blended and the paste was mixed with distilled water and then dispersed in a sodium chloride (4%) solution to separate proteins from the starch. The starch obtained in a separating funnel was then dried at 45°C for 48 hrs in an electric oven (Memmert, Germany).

The dried product was crushed and sieved under a 250 μ m mesh size.

Chemical composition of starch: The ash, lipid and protein contents were determined using the AOAC (1980) methods. Lipids were extracted by using Soxhlet apparatus and protein was determined according to Kjeldhal method using the conversion factor of 6.25. Amylose and amylopectin contents were carried out following the method described by Jarvis *et al.* (1993). Defatted starch (0.1 g) was dissolved in 5 mL of potassium hydroxide (1 N) solution. The suspension was thoroughly mixed and 5 mL of HCl (1N) solution were added. The mixture was boiled in water bath for 15 min and the volume was adjusted to 10 mL. After centrifugation at 3000 g for 10 min, the supernatant was used for determination of amylose and amylopectin. For

this, 0.05 mL of the supernatant was introduced in a test tube and 4.85 mL of distilled water, following by 0.1 mL of iodine reagent were added. The mixture obtained was left to stand for 10 min and the absorbance was read at 580 nm and 720 nm, respectively by using a spectrophotometer (Helios, Omega, UK). Standard curves of amylose and amylopectin were used as references.

Starch granule morphology: Isolated starch granules were mounted on the surface of a brass disk using double-sided adhesive silver-tape, coated with gold/palladium (60/40) and viewed under a scanning electron microscope (FEG Supra 40 VP Zeiss 2008). The diameter distribution was determined on 500 granules using a calibrated micrometer.

Solubility and swelling power: Swelling power and solubility of the starch were determined according to the method described by Leach *et al.* (1959).

Paste clarity: The paste clarity of starch samples was carried out as described by Zheng *et al.* (1998). Aqueous dispersions (1%) of starch were boiled at 100°C and constantly shaken for 30 min. The paste was cooled at ambient temperature and stored at 4°C for 4 weeks. The transmittance was measured at 650 nm every week by using a spectrophotometer (Helios, Omega, UK).

Freezing stability: Stability of starches samples to freezing was determined according to Eliasson & Kim (1992). Ten (10) g of paste sample were conditioned in a

plastic tube at -15°C and 4°C, respectively for 4 weeks. Freezing stabilities were performed every week by measuring the percentage of water expelled after centrifugation at 5000 g for 30 min.

In vitro digestibility:

Preparation of the enzyme solution: The digestive juice of the snail *Archachatina ventricosa* was collected according to the method described by Colas (1977). Snails were left fasting for three days and then the shell was carefully broken. The digestive tract was isolated and the coloured brown juice was collected. This digestive juice was centrifuged at 10,000 g for 15 min at 4 ° C (using a refrigerated centrifuge ALRESA). the supernatant filtered through cotton wool was refrigerated and used as the crude enzyme extract.

Enzymatic digestibility: An aliquot (2.5 mL) of starch suspension (1%; w/v) was mixed with 2.5 mL of sodium acetate buffer (100 mM pH 5.0) and 1.25 mL of crude enzyme extract. The mixture was incubated at 37°C in water bath for 5 h. Reducing sugars obtained by enzymatic hydrolysis were determined according to Bernfeld (1955) method.

Statistical analysis: All the experiences were performed in triplicate and the data were analyzed using EXCELL and XLSTAT version 2007. Differences between means were evaluated by Duncan's test. The significance level was set at p value < 0.05.

RESULTS

Starch content and chemical analysis: The yield of extracted maize starches was high and about 98.86% dry matter. The chemical composition of maize starch is given in Table 1. The ash were in small quantities in starches with values ranging from 0.08 to 0.1 ± 0.00% and there is no significant difference (p> 0.05) between values obtained for the four varieties. These starches showed fat and protein contents respectively in the range of 0.53 ± 0.05 to 0.66 ± 0.057 and 0.35 ± 0.06 to 0.39 ±

0.06. Concerning the fat of starches, QPM varieties do not show statistically significant differences (p> 0.05) between them, as well as for ordinary varieties. As against this, a significant difference (p<0.05) between the fat content of QPM and ordinary. As regard amylose and amylopectin, their content in starches studied, vary respectively 24.82 ± 0.11 to 25.94 ± 0.45 and 74.06 ± 0.45 to 75.18 ± 0.11.

Table 1: Chemical composition of QPM and ordinary maize starches (%dry basis)

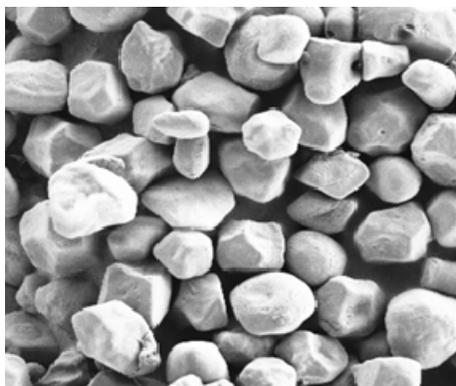
	White QPM	White ordinary	Yellow QPM	Yellow ordinary
Protein	0.37 ± 0.06 ^{ab}	0.39 ± 0.06 ^a	0.35 ± 0.06 ^b	0.38 ± 0.06 ^{ab}
Fat	0.53 ± 0.05 ^b	0.63 ± 0.05 ^a	0.57 ± 0.05 ^{ab}	0.66 ± 0.05 ^a
Ash	0.08 ± 0.00 ^a	0.10 ± 0.00 ^a	0.10 ± 0.00 ^a	0.10 ± 0.00 ^a
Amylose	25.11 ± 0.04 ^c	25.45 ± 0.23 ^b	24.82 ± 0.11 ^d	25.94 ± 0.45 ^a
Amylopectin	74.90 ± 0.04 ^{ab}	74.55 ± 0.23 ^b	75.18 ± 0.11 ^a	74.06 ± 0.45 ^c

Data are represented as means ± SEM (n=3). Mean with different letters in the same row are statistically different (p < 0.05) according to Duncan's test.

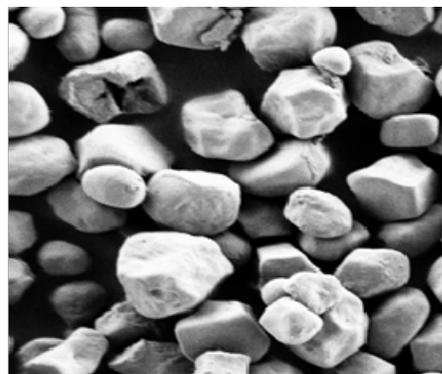
Morphological properties and particle size analysis:

The starch granules shapes and diameters measured by scanning electron microscopy are shown in Figure 1. The intervals and the mode of starch granule size distribution are given by Figure 2. The majority of starch granules were characterized by polyhedral and irregular shapes and only a small minority had spherical shapes. The particle size of the four starches (Figure 2) varied from 4.75 to 22.79 μm for white QPM; 5.28 to 22.62 μm for

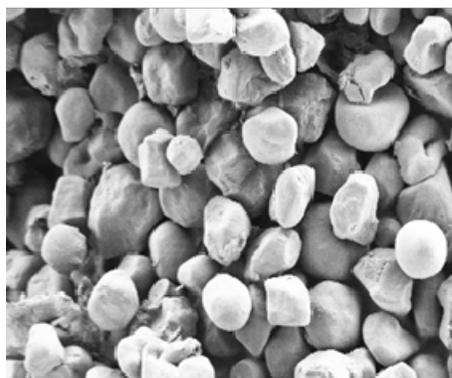
white ordinary maize; 5.98 to 22.88 μm for yellow QPM and 5.63 to 21.47 μm for yellow ordinary maize. The distribution of starch granules of both ordinary maize and QPM was unimodal. The principal components showed an average granule size of approximately $13.61 \pm 3.55 \mu\text{m}$ for white ordinary, $13.27 \pm 2.99 \mu\text{m}$ for yellow QPM, $14.07 \pm 3.20 \mu\text{m}$ for white QPM and $13.39 \pm 2.92 \mu\text{m}$ for yellow ordinary maize starch.



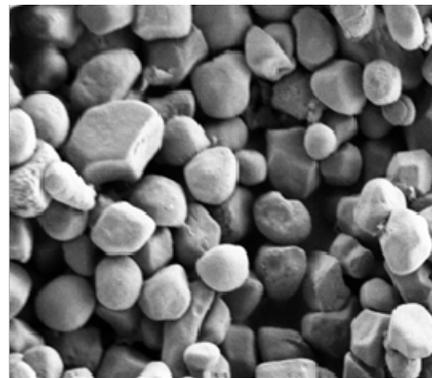
(a)



(b)



(c)



(d)

Figure 1: Scanning electron micrographs of maize starches (Gx400).

Legend: (a) white QPM (b) white ordinary (c) yellow QPM (d) yellow ordinary

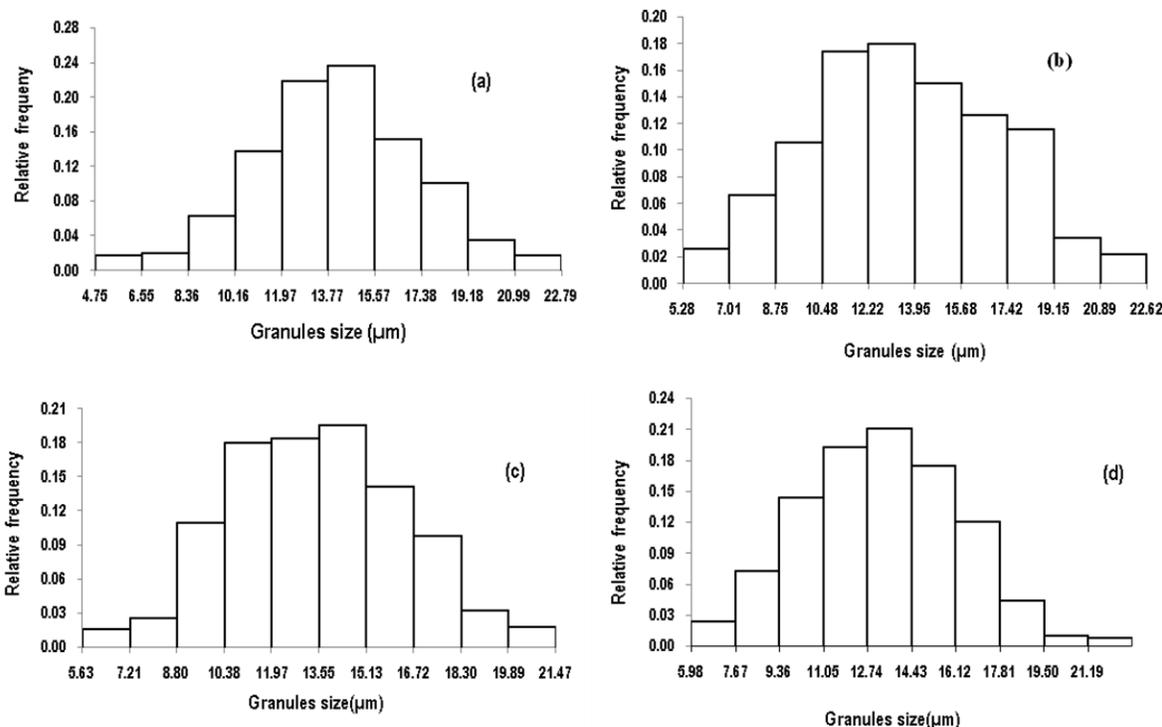


Figure 2: Frequency distribution of native starch granules average diameters

Legend: (a) white QPM ; (b) white ordinary; (c) yellow ordinary; (d) yellow QPM maize

Solubility and swelling power: The solubility and swelling power of studied maize starches at different temperatures are shown in Figure 3. Solubility of all maize starches increased with temperature and yellow QPM starch showed the highest solubility ($19.56 \pm 0, 12\%$) when heated at 95°C . Swelling power significantly increased exponentially with temperature in the range of

$55\text{-}95^\circ\text{C}$. Compared with ordinary maize, starches from white and yellow QPM were characterized by highest values of swelling power in the range of temperatures above. These values were $19.57 \pm 0, 94$ and $20.39 \pm 0, 50$ g/g at 95°C for yellow and white QPM starches, respectively.

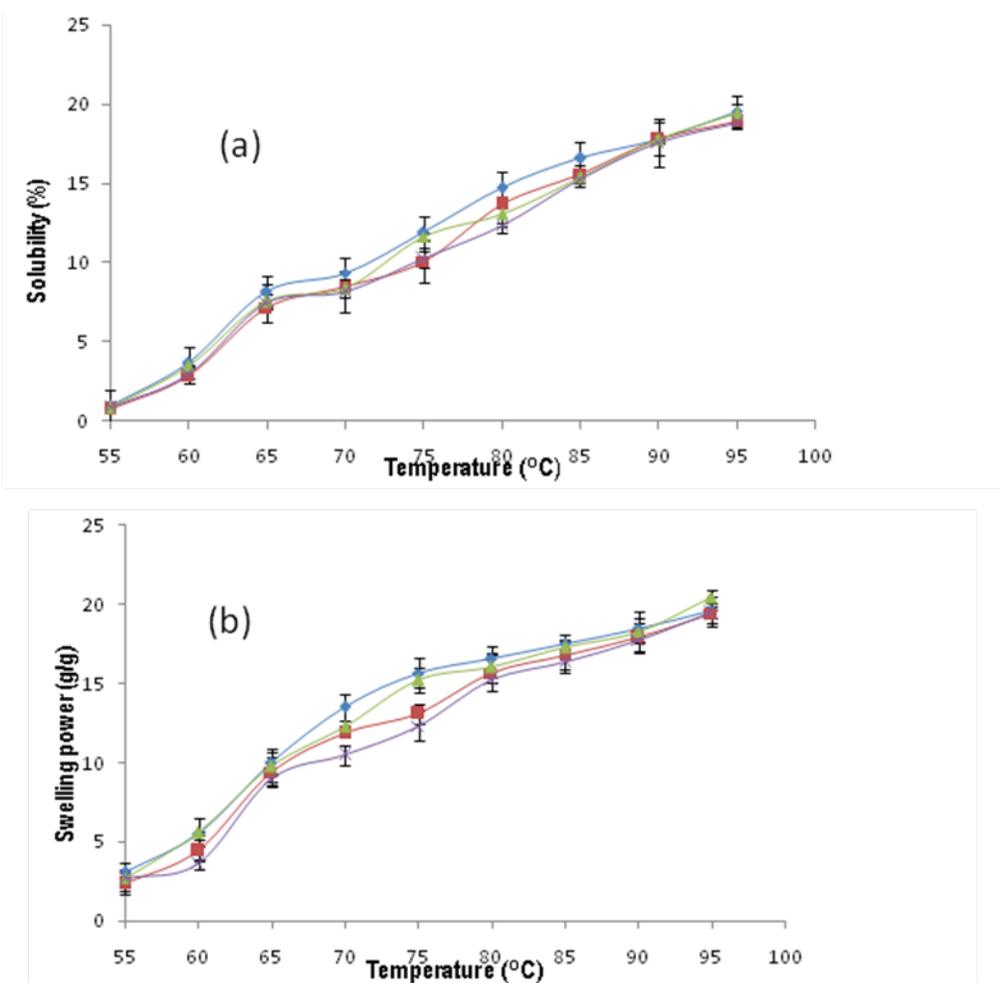


Figure 3: Solubility (a) and (b) swelling power (b) of maize starches
 Legend: ◆ yellow QPM ■ yellow ordinary maize ▲ white QPM × white ordinary maize

Syneresis and paste clarity: Paste syneresis of maize starches at -15°C and 4°C is shown in the Figure 4 while paste clarity of studied starches is illustrated in Figure 5. The syneresis of all maize starches increased during the first week under refrigeration and freezing conditions. After this period, relatively stable values of syneresis were observed for each studied starch. The percentage of water expelled from white QPM starch was $40.39 \pm 0, 72$ % at -15°C and $37.05 \pm 0, 5$ % at 4°C after the first week

of storage. Yellow QPM starch was characterized by syneresis values of $40.09 \pm 0, 3$ % at -15°C and $36.92 \pm 0, 2$ % at 4°C after the first week of storage. Both white and yellow QPM starches had the lowest values of syneresis compared to those of ordinary maize. The paste clarity of all starches (Figure 5) decreased quickly during the first week and then stabilized at about 0.74 to $1 \pm 0, 01$ %.

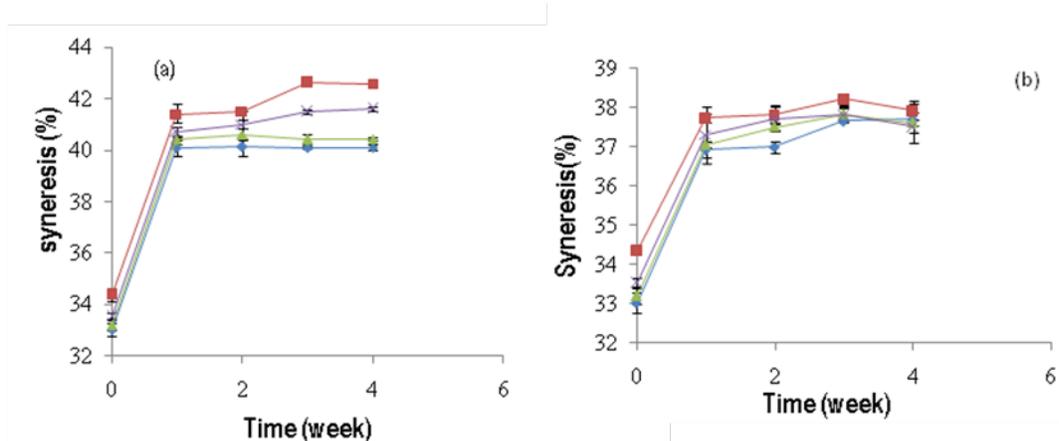


Figure 4: Syneresis and freeze thaw stability study of native maize starches of at -15°C (a) and 4°C (b).
 Legend: ♦ yellow QPM ■ yellow ordinary maize ▲ white QPM × white ordinary maize

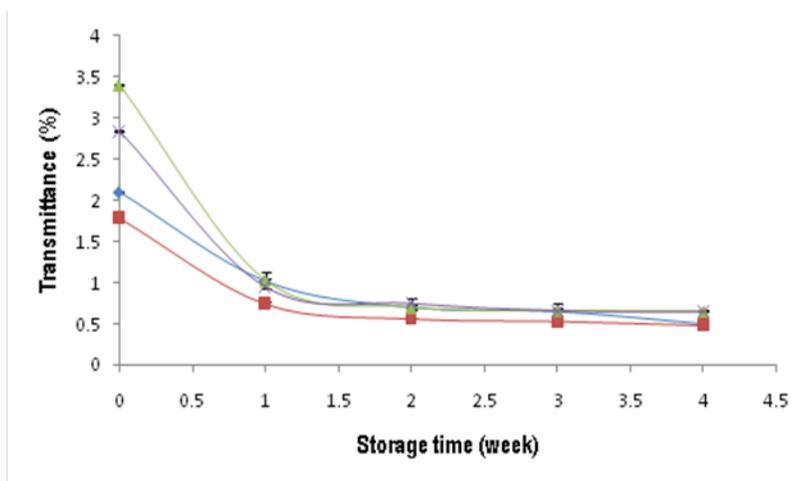


Figure 5: Paste clarity of maize starches at 4°C
 Legend : ♦ yellow QPM ■ yellow ordinary maize ▲ white QPM × white ordinary maize

Starch digestibility: Figure 6 shows results of gastric enzymatic digestion on maize paste starches. There was no difference about the kinetic of different paste starches

digestibility. Yellow QPM starch showed better in vitro digestibility after 60 min of enzymatic treatment.

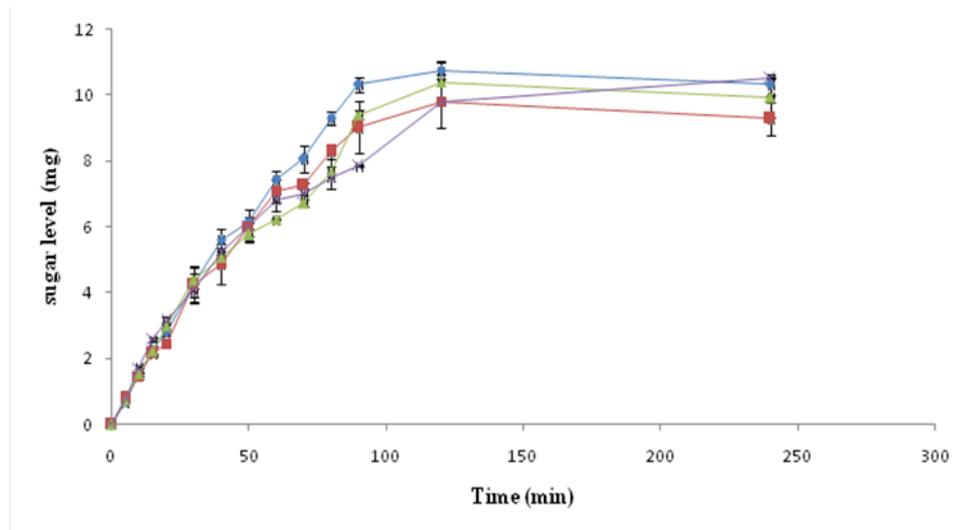


Figure 6: Kinetics of enzymatic hydrolysis of maize starches

Legend : ◆ yellow QPM ■ yellow ordinary maize ▲ white QPM × white ordinary maize

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DISCUSSION

The ash content of white and yellow QPM starches are below the limit of 0.5% recommended for grade industrial starches (Bradley, 1976). In addition, the lipid content of these starches was within the range of 0.1 to 1.4% reported in cereal starches (Abdel-Ail *et al.*, 2002). The relatively low values reported for ash, lipid and protein contents indicate a high purity of white and yellow QPM starches and these properties met the worldwide industrial standards (Swinkels, 1985; Thomas & Atwell, 1997). Amylose content of white and yellow QPM starches is approximately two-fold lower than that (47-50%) of cassava starch (Nuwamanya *et al.*, 2011). In view to the high retrogradation tendency of cassava and wheat starches, those extracted from white and yellow QPM could be suitable in bread making and other food application due to their potential extensibility (Soh *et al.*, 2006). As concern the morphological characteristics, QPM starch granules have the same morphology compared with ordinary maize. This finding may confer to white and yellow QPM starches, suitability for the manufacture of adhesives and these starches could be used in the cosmetic industry (Singh *et al.*, 2003).

Swelling power and solubility are usually used to assess the extent of interaction between starch chains, within the amorphous and crystalline domains of the starch granule (Ratnayake *et al.*, 2002). The highest solubility and swelling power of white and yellow QPM starch could be explained by granule size and amylose content. Indeed, starch with large granules swells rapidly when heated in water and water molecules are bonded to the free

hydroxyl groups of amylose and amylopectin by hydrogen bonds (Singh *et al.*, 2003). In addition, the low fat content in white and yellow QPM starches may have an increasing effect on the swelling power (Galliard & Bowler, 1987). Therefore, this highest swelling capacity could be an advantage for using these starches in food industry as thickeners (Jovin *et al.*, 2009).

Syneresis is a parameter, which represents the volume of water separated from the formed gel under storage at room temperature, refrigeration, and freezing. This parameter characterizes the starch stability to these temperatures (Salwa *et al.*, 2010). Syneresis in freeze-thawed gels is due to the rearrangement of amylose molecules in starch granules at reduced temperature, which acts to exclude water from the gel structure (Sandhu *et al.*, 2007). The lowest syneresis (highest resistance to freeze-thawing cycle) of yellow QPM starch may be attributed to the low amylose content or to the possible aggregation and crystallization occurring during the first hours of storage conditions (Singh *et al.*, 2006). The highest clarity of white QPM starch may be due to its lowest impurities (fat, ash, protein) contents and this property may be useful for application in food and textile industries where high clarity is required (Jyothi *et al.*, 2007). Hydrolysis of starch using amylolytic enzymes allows us to understand the total portion of starch that can be digested over a particular time. The high amylosis value (high digestibility) of white QPM starch may be linked to its low amylose content as suggested by Riley *et al.* (2006).

CONCLUSION

Starches extracted from white and yellow QPM cultivated in Côte d'Ivoire are practically similar in view to their physico-chemical and functional properties. Chemical characteristics of these starches have revealed low content in impurities and high content in amylose. Functional properties of starches from white and yellow

QPM have revealed highest swelling capacity, highest solubility and lowest syneresis. From these results, starches from white and yellow QPM present better properties than those of ordinary maize. Therefore starches extracted from white and yellow QPM could be more exploited in food and non-food industries.

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