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Changes of banana starch by chemical and physical modification

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Abstract

Detailed studies have been carried out on banana, *Musa* var. *valery* native and modified starches (gelatinized, phosphorylated, cross-linked phosphorylated and hydroxypropylated). Starch accounted for 33.8% of the banana dry weight of 99% of purity with compact granules irregularly shaped with elongated and spheroid forms (14–88 μ in width and 21–108 μ in length). Amylose content was 40.7%, λ_{max} of the iodine complex was 563 nm and *L* parameter of the CIELAB colour scale was low of 73.60. Hydroxypropyl banana starch had more than 50% higher water binding capacity than the native starch. Banana valery starch had fairly restricted swelling power and low solubility. Chemical modification produced improvement in those properties. Phosphoylated and hydroxypropylated banana starches showed improvement in clarity and phosphate starch has shown the best freeze–thaw stability. C type X-ray spectra was assigned to banana starch. Hydroxypropyl starch and starch phosphate have shown significant decrease in initial temperature of gelatinization.

Keywords: *Musa* var. *valery*; Native and modified starches

1. Introduction

The banana (genus *Musa*, AAA group), the largest herbaceous plant in the world, grown abundantly in many developing countries is considered to be one of the most important sources of energy for people living in the humid regions of many countries including México. It is also considered fourth on the list of the developing world's most important food crops, after rice, wheat and maize (Anon, 2002). Banana is a seasonal and highly perishable fruit and surplus fruits are often available year around. Due to high starch concentration (over 70% of dry weight), banana processing into flour and starch is of interest in view of a possibly important resource for food and other industrial purposes.

With around world 1000 types of banana, *Musa* var. *valery* is a small fruit size variety grown mostly in the southern states of México, mainly Tabasco. Due to a low commercial acceptance for export, it can be considered as a potential resource for industrial processing and starch production. Several studies on other varieties of banana starch were conducted mainly on isolation and partial characterization of physically modified starch (Lii, Chang, & Young, 1982; Bello-Perez, Agama-Acevedo, Sanchez-Hernandez, & Paredes-Lopez, 1999; Bello-Perez, Romero-Manilal, & Paredes-Lopez, 2000).

Native starches represent many disadvantages, thus limiting their wide application and industrial use. Functional properties of starches available on the commercial market, normally obtained from corn or other cereals, are often submitted for physical modification (mainly gelatinization) or for slight and relatively simple chemical modifications to fulfil needs of food and other industries. Modified food starches generally show better paste clarity and stability, increased resistance to retrogradation, and freeze–thaw stability (BeMiller, 1997).

The objective of this study was to characterize important physicochemical properties of native banana starch and the effect of physical treatment (gelatinization) and chemical modification (monoester or cross-linked phosphorylation and hydroxypropylation) on starch properties.

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2. Experimental

2.1. Plant material and starch isolation

Ready to harvest, green and unripened banana fruit (*Musa valery*) of 90 days after flowering was collected from the packaging plant 'Martin Banana' located 45 km west from Villahermosa, Tabasco state, México. The fruits were peeled and cut into 6 mm slices and each slice was cut into four pieces, which were rinsed immediately in sodium bisulfite solution (0.25 g/l) in 2:1 (v/w) proportion at 40 °C. Fruit pieces were macerated and blended at low speed (100 rpm) for 5 min. The resultant slurry was sieved through 100 mesh screen and washed three times or more until the waste solution was clean. Starch suspension was left overnight in refrigeration at 6 °C and washed with tap water and centrifuged at 3000 rpm for 15 min. The white sediment was dried at 40 °C in a convection oven for up to 48 h, ground in a mortar, passed through a 100 mesh screen, and stored at room temperature in sealed glass jars (AACC, 1983).

2.2. Starch proximate analysis

Moisture content was determined as weight loss after vacuum drying at 60 °C until constant weight. Ash, protein, fat and starch was determined according to AACC 08-01, 46-13, 30-25 and 76-13 methods, respectively (AACC, 1983).

2.3. Particle size analysis

A small amount of starch was covered with Au/Pd in shadower Fine Coat model Ion Sputter JFC-11000 in JSM-T20 Joel scanning electron microscope.

2.4. Amylose content

The amylose content was determined by modified enzymatic method (Paredes-Lopez, Bello-Perez, & Lopez, 1994).

2.5. Blue value (BV)

The blue value was determined by an iodometric method (Gilbert & Spragg, 1964). First, the maximum absorbance wavelength of starch–iodine complex was determined by scanning from 400 to 700 nm in a Perkin Elmer Lambda Bio 2.3 spectrophotometer.

2.6. Colour parameters

The most colour scale used in colour measurement of food ingredients including starch is the opponent CIELAB scale, measuring the degree of lightness (L), the degree of redness or greeness ($\pm a$) and the degree of yellowness or blueness ($\pm b$) (Mabon, 1993). For many food ingredients, L is used for darkness evaluation (100 = white and 0 = black). Samples of native and modified starches were subjected to colour analysis by five measurements in duplicate with the Minolta reflectance colorimeter Chroma Meter Cr-200 (Minolta Corp. Ramsey, NJ,). Results of each measurement are the means of three instrument repetitions. The instrument was calibrated against a standard white reference plate.

2.7. Preparation of starch cross-linked phosphate

200 ml of 45% starch suspension was mixed with 10 g of sodium sulphate and 4 g of trimetaphosphate (Lim & Seib, 1993). The pH of the suspension was adjusted to 9.5 by adding 10% aqueous hydrochloric acid or sodium hydroxide. The slurry was stirred for 1 h at room temperature, washed three times with distilled water and dried in an oven at 40 °C to 10–15% moisture and heated in an oil bath for phosphorylation for 2 h at 130 °C. After cooling to room temperature, the starch cake was washed many times by suspension in distilled water until unreacted phosphate was not detected and recovering the starch by centrifugation at 1500g for 10 min. Finally, the suspension was adjusted to pH 6.5, and the recovered starch was dried at 40 °C in a vacuum oven. The phosphate content was determined as described by Paschall (1964) and expressed in terms of molar substitution defined as mole of substitution per mole of anhydrous glucose unit. The results are means of three determinations.

2.8. Preparation of starch phosphate

The procedure was as mentioned above for cross-linked phosphate starch, but instead of using trimetaphosphate, 10 g of sodium tripolyphosphate was used. The phosphate content was determined as described by Paschall (1964) and expressed in terms of molar substitution defined as mole of substituent per mole of anhydrous glucose unit. The reported results are means of three determinations.

2.9. Preparation of hydroxypropyl starch

Starch was hydroxypropylated as described by Leegwater and Luten (1971). A sample of 100 g db of starch was suspended in 120 ml 0.1% NaOH with 15 g Na_2SO_4. After 10 min of reaction, 12 ml of propylene oxide was added and reaction was continued at 40 °C for 24 h with shaking. The starch suspension was then washed three times with distilled water. The pH was adjusted to 5.5 with 2 M HCl. The starch cake was washed with distilled water and dried at 40 °C in a vacuum oven. The hydroxypropyl content was determined by the spectrophotometric method (Johnson, 1969) and expressed in terms of molar substitution defined as moles of subsituent per mole of anhydro glucose unit. The results are means of three determinations.

2.10. Preparation of pregelatinized starch

A sample of 300 g of starch was suspended in 1 l of distilled water and heated to 80 °C for 15 min with slow mixing. Pregelatinized starch was placed into stainless steel tray in form of thin film (1–2 mm) and dried in a convection oven at 40 °C for 48 h, ground in a mortar to pass through a 100 mesh screen and stored at room temperature in sealed glass jars.

2.11. Swelling power and starch solubility

Starch five suspensions (1%w/w) was prepared in a flask and was heated to 50, 60, 70, 80, and 90 °C, respectively, for 30 min with shaking every 5 min and left for cooling to room temperature and centrifuged for 15 min at 3000*g*. The supernatant was decanted, and the residual volume was determined. The solid part was dried in an oven for 2 h at 130 °C.

2.12. Water retention capacity

Water retention was determined as described by Hallgren (1985). 5 g of starch and 100 ml of water were added to preweighed centrifuge tubes at room temperature and then heated to 50,60,70,80, and 90 °C for 15 min with shaking every 5 min period. Tubes were centrifuged at 3000*g* for 15 min, the supernatant was decanted, and the tubes were allowed to drain for 10 min at a 45° angle. The tubes were then weighed, and the gain in weight was used to calculate the water retention capacity.

2.13. Paste clarity

Starch suspension (4%w/w) in a screw cap tube was placed in boiling water for 30 min and thoroughly shaken every 5 min. After cooling to room temperature, some samples were refrigerated to 6 °C during 72 h and every 24 h the percent of transmitance at 650 nm was determined against water blank. The percent transmittance (%T) was determined at 650 nm against water blank in a Perkin Elmer Lambda Bio 2.3 spectrophotometer.

2.14. Paste freeze–thaw stability

Aqueous suspensions (5 ml) of starches (5%w/v) were rapidly heated 90 s to previously determined gelatinization temperature (Table 6) while under constant agitation. These suspensions were then held at 30 min before being cooled. The gels were subjected to cold storage at −20 °C for 18 h and then thawed to 28 °C for 6 h. The exuded water was determined gravimetrically by vortexing the thawed gels for 15 s followed by centrifugation at 3000 rpm for 10 min (Bello-Perez et al., 1999). The percentage of water separated after each freeze–thaw cycle was measured and expressed as the percentage of water separated. Ten freeze–thaw cycles were performed in total.

2.15. X-ray diffraction

X-ray diffractograms of strach powders were obtained with a Rigaku D-Max-2200 X-ray diffractometer (Rigaku Denki Co.). The scanning region of the diffraction angle was from 5 to 50° at 0.1° step size with a count time of 2 s. The starch sample was equilibrated in a 100% RH chamber for 24 h at room temperature.

2.16. Differential scanning calorimetry

Thermal transition of starch gelatinization was studied using a TA Instrument model 2010 differential scanning calorimeter (TA Instruments Ltd). Starch/water mixtures (water contents between 70 and 86% total wet basis) were used. The thermograms were acquired between 20 and 100 °C at a heating rate of 10 °C min^{-1}. An empty pan was used as a reference. Onset (T_0), peak (T_p) and conclusion (T_c) temperatures of the retrogradation endotherm were determined. The enthalpy of retrogradation (ΔH_R) was estimated by integrating the area between the thermogram and the base line under the peak and expressed in J g^{-1} of dry starch.

3. Results and discussion

Banana used for starch isolation was at the first stage of ripening as described by the Von Loesecke classification scale (Von Loesecke, 1950) according to the colour of the banana peel. The dry basis yield of starch was 33.8%. Bello-Perez et al. (1999) reported two yields of starch from macho and criollo banana 43.8 and 11.8%, respectively. This high difference is attributed to different ripening stages. Banana starch was of high purity (over 99%), it contained 0.22% of protein, 0.26% fat and 0.47% ash. Results of protein and fat are comparable with results of other authors (Kayisu, Hood, & Vansoest, 1981; Lii et al., 1982). Higher results of ash may be due to higher potassium and magnesium content in valery banana variety. Much higher results of protein, fat and ash were found in macho and criollo banana starch (Bello-Perez et al., 1999); that may have been the result of a different technique of starch purification. Molar substitution of starch cross-linked phosphate, starch phosphate and hydroxypropyl starch were 0.20, 0.16 and 0.017, respectively.

Scanning electron micrographs of banana starch showed compacted granules, irregularly shaped with elongated and spheroid forms (14–88 μ in width and 21–108 μ in length), very similar but larger than those found by Kayisu et al. (1981). Small particles of damaged starch granules appeared but the quantity was below 1% and it means that the technique of starch preparation did not cause significant damage to the granules.

Table 1
Water binding capacity of native and chemically modified banana starch submitted to heat from 50 to 90 °C (g of water retention g^{-1} of starch × 100)

Type of starch/temp	50 °C	60 °C	70 °C	80 °C	90 °C
Native	8.9	19.3	35.1	40.4	44.7
Phosphate	16.7	30.7	41.4	46.1	49.1
Cross-linked phosphate	9.6	19.6	34.7	42.7	44.7
Hydroxypropyl	11.5	29.8	48.8	60.8	68.8
Pregelatinized	40.3	42.8	42.9	43.1	43.0

Means of three replicates.

The λ_{max} of the iodine complex wiyj banana valery starch was 563 nm, a result quite different, with macho (583 nm) and criollo (589 nm) banana starches (Bello-Perez et al., 1999). In the study of the potato amylose and amylopectin complex with iodine, McGrance, Cornell, and Rix (1998) have shown extremely broad peaks in visible spectra. The authors have determined maximum absorbance for pure amylopectin at 552 nm and at 636 nm for pure amylose. Amylose content in banana valery starch was 40.7% and was different for cavendish banana starch (19.5%) (Ling, Osman, Fernandes, & Ames 1982), macho (18%) and criollo (87%) banana starches starch (Bello-Perez et al., 1999),

Compared to many starches, *L* result of banana starch was low (73.60) due to slight yellowness and darkness. The process of chemical modification has shown more darkness, lowering *L* result significantly to 66.30 for starch phosphate, 61.09 for starch cross-linked phosphate, 57.26 for hydroxypropyl starch and 48.21 for gelatinized starch, respectively. This means that banana valery modified starches cannot be offered for clear starchy products. Results of chroma (colour intensity) underwent the following changes: from 15.05 for native starch decreased to 11.92 for starch phosphate, 13.68 for starch cross-linked phosphate, but increased to 18.48 for hydroxypropyl starch and 23.27 for pregelatinized starch.

Results of water binding capacity of native and chemically modified starches submitted to heating from 50 to 90 °C are shown in Table 1. Chemical modification improved starch water binding because the hydrophilic groups were incorporated. At the lowest temperature (50 °C), the highest increase in water binding was observed in phosphate modified starch. With temperature increase up to 90 °C, water binding capacity has a tendency to increase and the highest results was obtained with hydroxypropyl starch. At 90 °C, this modified starch has shown over 50% higher water binding capacity than native starch. Native banana valery starch has shown almost the same water binding capacity as macho and criollo banana starches (Bello-Perez et al., 1999),

Table 2
Swelling power of native and chemically modified banana starch submitted to heat from 50 to 90 °C (g water g^{-1} dry starch)

Type of starch/temp	50 °C	60 °C	70 °C	80 °C	90 °C
Native	1.8	2.2	2.3	7.8	8.7
Phosphate	3.3	6.1	6.7	9.0	9.8
Cross-linked phosphate	1.9	3.6	5.9	9.0	9.0
Hydroxypropyl	2.3	3.2	9.0	12.4	13.8
Pregelatinized	8.0	8.5	8.6	8.6	8.6

Means of three replicates.

Table 3
Solubility of native and chemically modified banana starch submitted to heat from 50 to 90 °C (g water g^{-1} dry starch × 100)

Type of starch/temp	50 °C	60 °C	70 °C	80 °C	90 °C
Native	1.8	2.2	2.9	7.8	8.7
Phosphate	3.3	5.1	6.1	9.0	9.8
Cross-linked phosphate	1.9	2.1	2.5	8.8	9.0
Hydroxypropyl	2.3	3.2	10.0	13.6	13.8
Pregelatinized	8.0	8.6	8.7	8.7	8.6

Means of three replicates.

The swelling power of native and chemically modified starch was measured at 10 °C intervals from 50 to 90 °C (Table 2). The results indicate that banana valery starch has fairly restricted swelling power but not to the extent of legume pregelatinized starches. Starch and starch phosphate and cross-linked phosphate have shown small improvement in swelling power except, but hydroxypropyl starch did not.

Native banana starch has shown low solubility and it was improved by chemical modification or simple pregelatinization (Table 3). Changes were found in pregelatinized starch due to temperature increase from 60 to 90 °C. Starch solubility was increased by chemical modification, mainly hydroxypropyl substitution and it may be due to the repulsion between positively charged modified groups on the starch molecules, lowering the intermolecular bonding forces (Sitohy, El-Saadany, & Ramadan, 2000).

Differences in paste clarity were observed between starches (Table 4). Phosphate and hydroxypropyl starches have shown improvement in paste clarity due to chemical modification, but cross-linked phosphate starch gave lower paste clarity than native starch. The changes to granular and molecular structures induced by hydroxypropylation and phosphorylation facilitated water penetration and absorption on the starch granules, which ultimately led to more swelling of starch and resulting in more transmittance of light (Craig, Maningat, Seib, & Hoseney, 1989). However, no such trend can be observed with all types of starches, for

Table 4
Effect of storage on paste clarity of native and chemically modified banana starch

Type of starch/days	0	24	48	72
Native	1.2	0.7	0.4	0.3
Phosphate	1.8	1.5	1.3	1.2
Cross-linked phosphate	0.7	0.6	0.4	0.4
Hydroxypropyl	2.3	2.0	1.9	1.8
Pregelatinized	13.3	11.7	11.7	11.1

Table 5
Effect of freeze–thaw cycles on percentages of separated water

Cycle/ starch	Native	Phosphate	Cross-linked phosphate	Hydroxy-propyl	Pregelatinized
1	2	0	10	16	17
2	5	0	11	60	22
3	8	0	12	62	23
4	25	0	13	64	23
5	33	5	15	66	23
6	39	6	16	67	24
7	47	10	17	68	24
8	49	10	19	68	24
9	50	11	20	69	25
10	59	13	22	70	27

example amaranth starch (Bhandari & Singhal, 2002). In this study, all modified starches have shown a tendency to decrease in clarity during storage of up to 72 h of at ambient temperature.

After three freeze–thaw cycles, native banana starch presented poor freeze–thaw stability (Table 5). Phosphate starch has shown the best stability and no syneresis was observed in four cycles. After ten cycles, only 10% of syneresis was determined.

The wide-angle X-ray (Fig. 1) diffractogram exhibited strong diffraction peaks at 15.3 and 17.2° ($2\theta/\theta$), and one very broad peak from 22 to 24°. The typical peak for potato starch and other B type starches at 5.7° was absent. The spectrum was somewhat similar to that of waxy corn starch (A type) but the typical peak at 18° was absent indicating that C type X-ray spectra should be assigned (Hizukuri, 1961). Gidley (1987) suggested that the C type is not true crystalline polymorph but rather a mixture of A and B polymorphs. Cairns, Bogracheva, Ring, Hedley, and Morris (1997) supported this argument by calculating the proportion of A and B polymorphs in the C type pea starch by computing matching X-ray spectra from a mixture of corn (A) and potato (B) diffractograms.

The gelatinization process of starch can be simply monitored by differential scanning calorimetry and

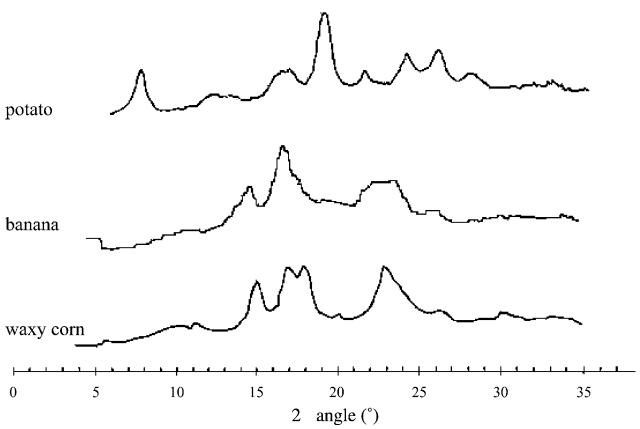

Fig. 1. The wide-angle X-ray diffractogram of potato, banana and waxy corn starches.

Table 6
Differential scanning calorimetry of native and chemically modified banana starch

Type of starch	T_0	T_p	T_c	$T_c - T_o$	ΔH (J g^{-1})
Native	69.5	74.6	81.2	12.7	5.2
phosphate	62.9	69.5	73.8	10.9	49.7
Cross-linked phosphate	71.7	76.0	81.3	9.6	25.3
Cross-linked phosphate	60.2	72.5	80.5	20.3	98.6

the gelatinization temperature obtained from thermograms was defined as the initial temperature (T_0), peak temperature (T_p), and completion temperature (T_c) (Table 6). The gelatinization temperature of valery banana starch (69.5 °C) was in the range of 62.3–72.0 °C obtained for different varieties of banana flour (Cairns et al., 1997). Lii et al. (1982) investigated some physico-chemical properties of banana starches due to ripening stage and for stage 1, temperatures 75, 77.5 and 80 °C for T_0, T_p and T_c, respectively, were found, to be a little bit lower than our results, but gelatinization enthalpy was almost the same (5.0 J g^{-1}). Higher results of ΔH were obtained (10.8–13.3) by other authors, probably due to impurities in banana flour (Da Mota, Lajolo, Ciacco, & Cordenunsi, 2000). Hydroxypropyl starch and starch phosphate have shown significant decrease in initial temperature of gelatinization and all chemically modified starches have shown significant enhancement in heat gelatinization.

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