

Structural, morphological, and physicochemical properties of acetylated high-, medium-, and low-amylose rice starches

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ABSTRACT

The high-, medium-, and low-amylose rice starches were isolated by the alkaline method and acetylated by using acetic anhydride for 10, 30, and 90 min of reaction. The degree of substitution (DS), the Fourier-transformed infrared spectroscopy (FTIR), the X-ray diffractograms, the thermal, morphological, and pasting properties, and the swelling power and solubility of native and acetylated starches were evaluated. The DS of the low-amylose rice starch was higher than the DS of the medium- and the high-amylose rice starches. The introduction of acetyl groups was confirmed by FTIR spectroscopy. The acetylation treatment reduced the crystallinity, the viscosity, the swelling power, and the solubility of rice starch; however, there was an increase in the thermal stability of rice starch modified by acetylation.

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1. Introduction

Starch is composed of amylose and amylopectin molecules and the ratio between both molecules varies according to the botanical origin of starch. Starch is the major constituent of rice grains and is considered an important ingredient that has been used in food preparation (Bao, Kong, Xie, & Xu, 2004; Blazek & Gilbert, 2011). Due to the wide range of amylose levels, rice starch has been used as an ingredient in various food and industrial products, such as desserts, bakery products, and alternatives to fats (Puchongkavarin, Varavinit, & Berghaller, 2005).

Native starches do not always have the desired properties for certain types of processing. In order to achieve suitable functionalities for various industrial applications, starch has been modified by different methods. Basically, there are four kinds of modifications: chemical, physical, genetic, and enzymatic (Kaur, Ariffin, Bhat, & Karim, 2012). Chemical modifications can promote structural changes and introduce new functional groups that affect the

physical and chemical properties of starches (Sandhu, Kaur, Singh, & Lim, 2008).

Acetylation converts the hydroxyl groups of the glucose monomers into acetyl groups (Graaf, Broekroelofs, Janssen, & Beenackers, 1995). The acetylated starches are classified into low, intermediate, or high degrees of substitution (DS). Acetylated starches with a low DS (0.01–0.2) may function as film-forming, binding, adhesion, thickening, stabilizing, and texturing agents, and are widely used in a large variety of foods including baked goods, canned pie fillings, sauces, retorted soups, frozen foods, baby foods, salad dressings, and snack foods. Acetylated starches with intermediate DS (0.2–1.5) and high DS (1.5–3) have high solubility in acetone and chloroform and, thus, have been reported as a thermoplastic material (Luo & Shi, 2012).

Acetylation may be performed to improve the physical, chemical, and functional properties of the starch (Xu, Miladinov, & Hanna, 2004) and has been widely studied by several researchers (Bello-Pérez, Agama-Acevedo, Zamudio-Flores, Mendez-Montealvo, & Rodriguez-Ambriz, 2010; Diop, Li, Xie, & Shi, 2011; Garg & Jana, 2011; Huang, Schols, Jin, Sulmann, & Voragen, 2007; Mbougung, Tenin, Scher, & Tchiégang, 2012). The changes introduced by acetylation depend on the botanical source, the degree of substitution, the ratio between amylose and amylopectin, and the molecular

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structure of the starch. The number of acetyl groups incorporated into the starch molecule during acetylation and the efficiency of the reaction depend on the type of reagent, reagent concentration, pH of reaction, presence of catalyst, reaction time, botanical origin, and size and structure characteristics of the starch granules (Huang et al., 2007; Huber & BeMiller, 2000).

Several researchers have reported the effects of acetylation on potato, corn, and pea starch properties (Chen, Li, Li, & Guo, 2007; Elomaa, 2004; Graaf et al., 1995; Xu & Hanna, 2005; Huang et al., 2007). A recent study performed by Luo and Shi (2012) showed effects of acetylation on waxy, normal, and high-amylose maize starch properties. There are few studies about the effects of acetylation of starches with a wide range of amylose contents. Sodhi and Singh (2005) studied the characteristics of acetylated starches from different rice cultivars with an amylose content between 7.83% and 18.86%; however, this study did not consider the effects of acetylation reaction time on starch properties. The aim of this study was to evaluate the effects of acetylation with different DS on FTIR spectroscopy, X-ray diffraction, thermal, morphological, and pasting properties, swelling power and solubility of high-, medium-, and low-amylose rice starches.

2. Materials and methods

2.1. Material

Rice grains of cultivars IRGA 417 (high-amylose), IRGA 416 (medium-amylose), and Motti (low-amylose), with amylose contents of 32%, 20%, and 8%, and purity of 99.4%, 99.5% and 99.1%, respectively, were used. Rice samples were dehulled, polished, and ground in order to obtain rice flour. Rice starch was isolated with 0.1% NaOH as described by Wang and Wang (2004). Rice flour was soaked in 0.18% NaOH at a 1:2 (w/v) ratio for 18 h. Then it was blended, passed through a 63 μ m screen, and centrifuged at 1200 $\times$ g for 5 min. The soft top layer was carefully removed, and the underlying starch layer was re-slurried. The starch layer was then washed twice with 0.18% NaOH and centrifuged. The starch layer was washed with distilled water and centrifuged. The starch was then re-slurried and neutralized with 1.0 M HCl to a pH of 6.5 and centrifuged. The neutralized starch was washed with distilled water three times and dried at 40 °C until 7% moisture content was achieved.

2.2. Starch acetylation

The high-, medium-, and low-amylose rice starches were acetylated according to the method described by Mark and Mehlretter (1972), with some modifications. Starch (200 g) was dispersed in 600 ml acetic anhydride in a closed reactor 2000 rpm for 5 min (RW 20, IKA, Germany). Afterwards, 20 g of 50% NaOH in water were added to the slurry and the temperature was adjusted to 90 °C for 15 min. The reaction was performed for three different times: 10, 30, and 90 min. When the time of reaction from each treatment was achieved, the temperature was reduced to 25 °C and 300 mL of 92.6° Gl ethanol was added to the slurry in order to precipitate starch. The material was centrifuged at 3000 $\times$ g for 10 min, suspended in alcohol for four times, and finally dried in an oven at 40 °C for 16 h.

2.3. Determination of acetyl percentage (Ac%) and degree of substitution (DS)

The percentage of acetyl groups (Ac%) and the degree of substitution (DS) of the acetylated starches were determined by the titration method described by Wurzburg (1964). Acetylated starch (1 g) was mixed with 50 ml of 75% ethanol in distilled water. The

250 ml flask containing the slurry was covered with aluminum foil and placed in a water bath at 50 °C for 30 min. The samples were then cooled and 40 ml of 0.5 N KOH were added. The slurry was kept under constant stirring at 200 rpm for 72 h. After this period, the alkali excess was titrated with 0.05 N HCl, using phenolphthalein as indicator. The solution was left to stand for 2 h and then any additional alkali, which may have leached from the sample, was titrated. A blank, using the original unmodified starch, was also used.

$$\text{Ac \%} = \frac{[\text{blank} - \text{sample}] \times \text{molarity of HCl} + 0.043 \times 100}{\text{sample weight}} \quad (1)$$

Blank and sample titration volumes were expressed in mL, sample weight was expressed in g. DS is defined as the average number of sites per glucose unit that possess a (Whistler & Daniel, 1995).

$$\text{DS} = \frac{162 \times \text{acetyl \%}}{4300 - [42 \times \text{acetyl \%}]} \quad (2)$$

2.4. Fourier transform infrared (FTIR) spectroscopy

The infrared spectra of the native and acetylated starches were obtained using a Fourier transform infrared (FTIR) spectrometer Prestige-21, Shimadzu, in the region of 4000–400 cm^{-1} . Pellets were created by mixing the sample with KBr at a ratio of 1:100 (sample:KBr). Ten readings were collected at a resolution of 4 cm^{-1} .

2.5. X-ray diffraction

X-ray diffractograms of the native and acetylated starches were obtained with an XRD-6000 (Shimadzu, Kyoto, Japan) diffractometer. The scanning region of the diffraction ranged from 5 to 40°, with a target voltage of 30 kV, a current of 30 mA, and a scan speed of 1° min^{-1} . The relative crystallinity (RC) of the starch granules was calculated as described by Rabek (1980) using the equation $\text{RC (\%)} = (\text{Ac}/(\text{Ac} + \text{Aa})) \times 100$, where Ac and Aa are the crystalline and amorphous areas, respectively.

2.6. Thermal analysis

Thermal analysis of the starch samples was performed in a TG-DTA apparatus (DTG model 2010, TA Instruments, New Castle, USA). Change in sample weight against temperature (thermogravimetric analysis, TG) and heat released or absorbed in the sample because of exothermic or endothermic activity in the sample (differential thermal analysis, DTA) were measured. Samples (4–8 mg) were heated from 30 °C to 600 °C at a heating rate of 10 °C/min. Nitrogen was used as purge gas at a flow rate of 50 mL/min.

The gelatinization characteristics of starches were determined using differential scanning calorimetry (DSC) (DSC model 2010, TA Instruments, New Castle, USA). Starch samples (approximately 2.5 mg, dry basis) were weighed directly in an aluminum pan, and distilled water was added to obtain a starch–water ratio of 1:3 (w/w). The pan was hermetically sealed and allowed to equilibrate for one hour before analysis. The sample pans were then heated from 30 to 120 °C at a rate of 10 °C/min. An empty pan was used as a reference. The temperature at the onset of gelatinization (T_0), the temperature at peak (T_p), the temperature at the end of gelatinization (T_c) and the enthalpy (ΔH) of gelatinization were determined.

2.7. Morphology of the starch granules

Starch samples with 7% moisture content were initially suspended in acetone to obtain a 1% (w/v) suspension, and the samples were maintained in an ultrasound for 15 min to eliminate the presence of air bubbles. A small quantity of each sample was spread directly onto the surface of the stub and dried in an oven at 32 °C

Table 1

Percentage of acetyl groups (Ac%) and degree of substitution (DS) of high-, medium- and low-amylose rice starches acetylated under different reaction times.

Starches	Acetylation time (min)					
	10		30		90	
	Ac%	DS	Ac%	DS	Ac%	DS
High-amylose	6.17 ^c	0.24 ^c	10.22 ^c	0.42 ^c	16.10 ^c	0.72 ^c
Medium-amylose	9.23 ^b	0.38 ^b	10.75 ^b	0.45 ^b	17.80 ^b	0.81 ^b
Low-amylose	10.34 ^a	0.43 ^a	11.60 ^a	0.49 ^a	20.47 ^a	0.96 ^a

Results are the means of three determinations. Values accompanied by different letters in the same column statistically differ ($p < 0.05$).

for 1 h. Subsequently, all of the samples were coated with gold and examined in the scanning electron microscope under an acceleration voltage of 15 kV and magnification of 5000 $\times$.

2.8. Pasting properties

The pasting properties of the starch samples were determined using a Rapid Visco Analyser (RVA–4, Newport Scientific, Australia) with a Standard Analysis 1 profile. The viscosity was expressed in rapid visco units (RVU). Starch (3.0 g of 14 g/100 g wet basis) was weighted directly in the RVA canister, and 25 ml of distilled water was then added to the canister. The sample was held at 50 °C for 1 min, heated to 95 °C in 3.5 min, and then kept at 95 °C for 2.5 min. The sample was cooled to 50 °C in 4 min and then kept at 50 °C for 1 min. The rotating speed was maintained at 960 rpm for 10 s, and it was maintained at 160 rpm during the remaining process. Parameters including pasting temperature, peak viscosity, holding viscosity, breakdown, final viscosity, and setback were recorded.

2.9. Swelling power and solubility

The swelling power and solubility of the starches were determined as described by Leach, McCowen, and Schoch (1959). Samples (1.0 g) were mixed with 50 mL of distilled water in centrifugal tubes. The suspensions were heated at 90 °C for 30 min. The gelatinized samples were then cooled to room temperature and centrifuged at 1000 $\times$ g for 20 min. The supernatant was dried at 110 °C to a constant weight to quantify the soluble fraction. The solubility was expressed as the percentage of dried solid weight based on the weight of the dry sample. The swelling power was represented as the ratio of the weight of the wet sediment to the weight of the initial dry sample (deducting the amount of soluble starch).

2.10. Statistical analysis

Analytical determinations for the samples were performed in triplicate and standard deviations were reported, except for X-ray diffraction and thermal analysis, which were performed twice. A comparison of the means was ascertained by Tukey's test to a 5% level of significance using analysis of variance (ANOVA).

3. Results and discussion

3.1. Percentage of acetyl groups (Ac%) and degree of substitution (DS)

The acetylation of starch yielded different rice starch DS values depending on the amylose content and time of reaction (Table 1). Low-amylose content and long time of reaction resulted in the highest Ac% and, thus, the highest DS. The rice starches acetylated for 90 min of reaction showed higher DS than the starches acetylated

for 10 and 30 min of reaction. The low-amylose rice starch exhibited greater ability for the insertion of acetyl groups compared to medium- and high-amylose starches. Luo and Shi (2012) studied the characteristics of acetylated high-amylose, normal, and waxy maize starches, reporting similar results. These authors justified the greater ease of insertion of acetyl groups of the waxy starch compared to the high-amylose maize starch as being due to the greater extent of reaction sites in the waxy starch. Sodhi and Singh (2005) acetylated the starch from different rice cultivars and reported that the variation in DS among different rice starches may be due to the difference in intragranular packaging. They reported that the way in which the amylose chain is packed in amorphous regions as well as the arrangement of amylose and amylopectin chains could affect the chemical substitution reaction in the glucose units of starch macromolecules. However, the effects of starch acetylation as related to amylose content have not been explained.

The acylation of starch takes place by an addition–elimination mechanism (Xu et al., 2004). Each one of the three free hydroxyl groups of the starch shows different reactivity (Garg & Jana, 2011). The primary C(6)OH is more reactive and is acylated more readily than the secondary ones on C(2) and C(3) due to steric hindrance. This fact can justify the highest degree of substitution of the starch with low amylose content. Of the two secondary OH groups, C(2)OH is more reactive than C(3), mainly because the former is closer to the hemi-acetal and more acidic than the latter (Fedorova & Rogovin, 1963). Since C(6) is the most reactive, it has been the main reactive site for substitution of the hydroxyl groups by acetyl groups.

3.2. Fourier transform infrared (FTIR) spectroscopy

FTIR spectroscopy analysis was used to monitor changes in the structure of the starches promoted by acetylation by analyzing the frequency and the intensity of the peaks. Fig. 1 presents the FTIR spectra of native and acetylated high-, medium-, and low-amylose rice starches. There was no difference in the FTIR spectra of high-, medium-, and low-amylose rice starches.

The native and acetylated starches showed peaks at 3450 cm^{-1} , which is assigned to the vibration of O–H deformation, and at 2960 cm^{-1} , which can be attributed to C–H bond stretching (Diop et al., 2011). The acetylated high-, medium-, and low-amylose starches, at all reaction times, showed the introduction of the carbonyl group (C=O) of the esterified acetyl groups, being verified by the band at 1750 cm^{-1} (Fig. 1). Moreover there was a decrease in the intensity of the band at 1650 cm^{-1} in the acetylated starches compared to their respective native starches. The peak of starch at 1650 cm^{-1} was assigned as C–O–C stretching, which can be attributed to the water associated to starch molecules. The reduction of this band in acetylated starches is the result of lower affinity to water as compared with native starches. Luo and Shi (2012) also reported that acetylated starches have a hydrophobic character due to the insertion of acetyl groups in the starch chains.

3.3. X-ray diffraction

The X-ray diffractograms of native and acetylated rice starches are presented in Fig. 2. The native and acetylated rice starches showed diffraction patterns typical of A-type crystalline structure as defined by peaks at 2θ of 15°, 17°, 17.8°, 19°, and 23°. The crystallinity of the native starches followed the order: low-amylose > medium-amylose > high-amylose. The higher crystallinity of the low-amylose native starch is attributed to its higher amylopectin content. The acetylated rice starches showed a decrease in the intensities of the peaks compared to the native ones, with the exception of low-amylose starch acetylated for 90 min of reaction. Acetylation reduced the crystallinity of rice starches, and the lowest values of relative crystallinity were seen in acetylated

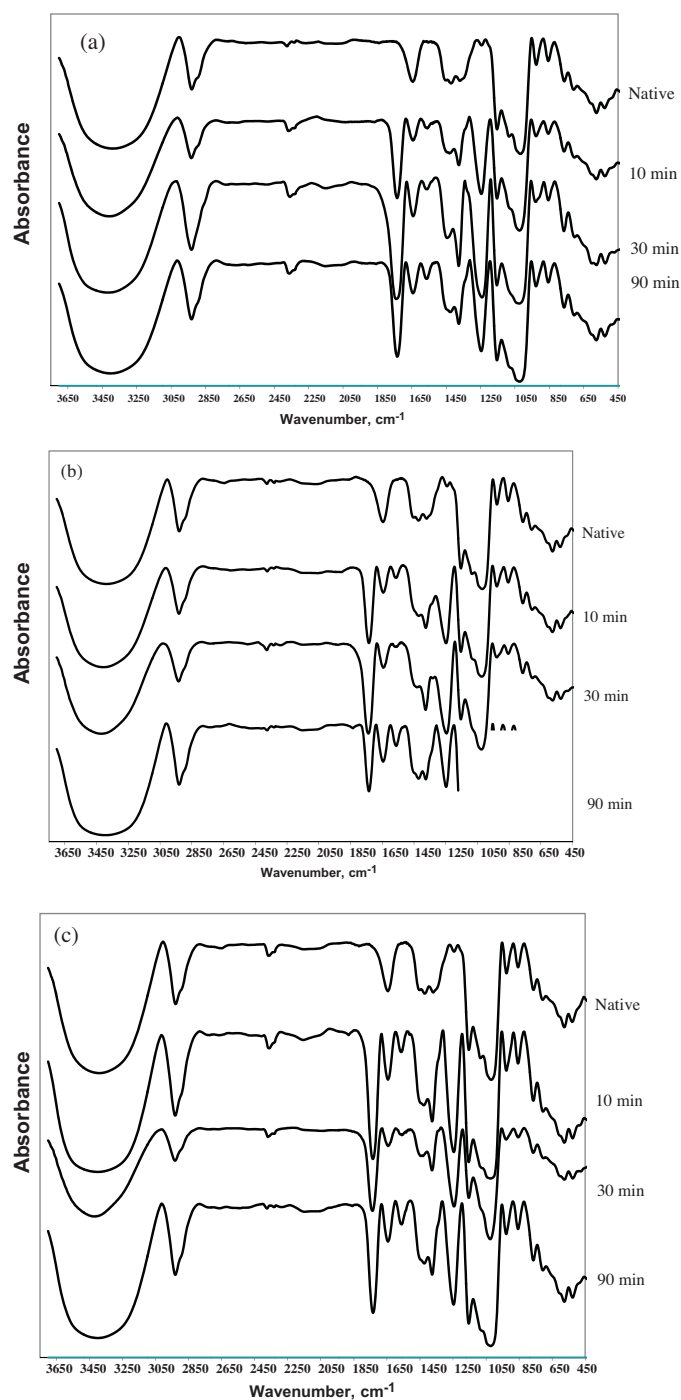


Fig. 1. FTIR spectra of native and acetylated rice starches. High-amylose starch (a), medium-amylose starch (b), and low-amylose starch (c).

starches with the highest DS. [Sha et al. \(2012\)](#) reported that, with the increase in the proportion in acetyl content of the rice starch, crystallinity became gradually lowered and the diffraction peak also reduced in turn. They described that the changes in the diffraction patterns indicated that the intermolecular hydrogen bonding interaction was damaged.

According to [Luo and Shi \(2012\)](#), acetylation reduces the formation of intermolecular hydrogen bonds, resulting in a low ordered crystalline structure of starch granules. These authors studied the acetylation of maize starches with varying DS, between 0.27 and 1.29, and reported that a destruction of crystalline structure occurred in high-amylose starch with 120 min of reaction. [Xu et al.](#)

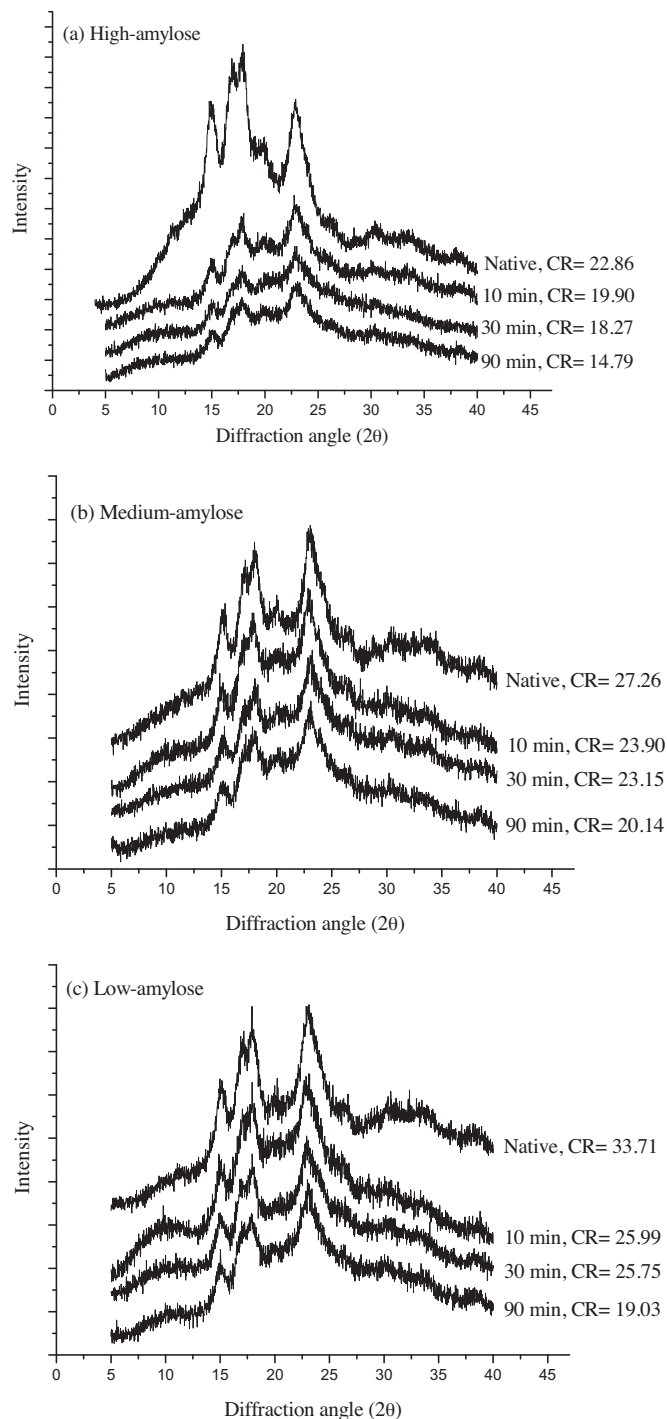


Fig. 2. X-ray diffraction pattern of native and acetylated rice starches. High-amylose starch (a), medium-amylose starch (b), and low-amylose starch (c).

(2004) also reported that the high-amylose maize starch, when acetylated with DS between 1.11 and 2.23, showed destruction in the ordered crystalline structures.

3.4. Thermal analysis

Thermogravimetric analysis (TGA) has been used in the evaluation of the thermal stability of materials and is considered one of the main methods for evaluating thermal properties of acetylated starches. The TGA curves showed two-stage weight loss for the studied starches, being the first stage around 40–125 °C and the

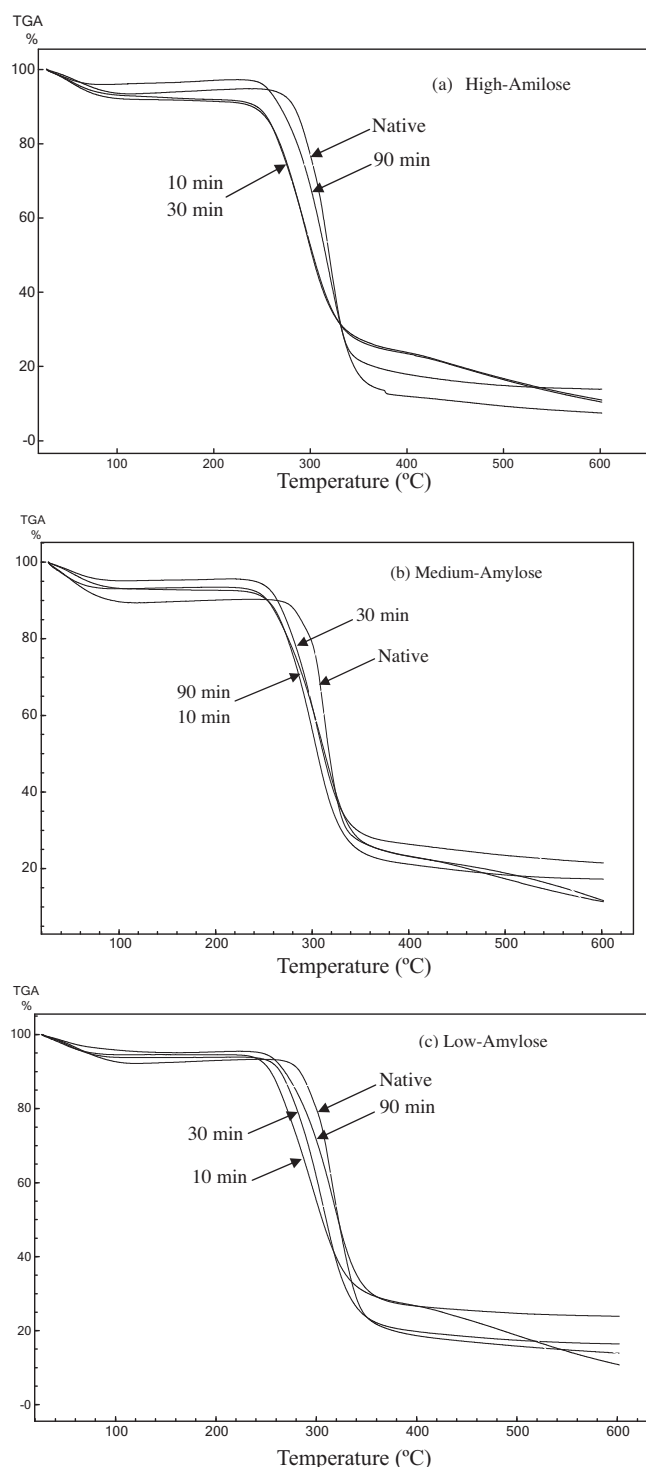


Fig. 3. Thermogravimetric analysis (TGA) curves of native and acetylated rice starches. High-amylose starch (a), medium-amylose starch (b), and low-amylose starch (c).

second one around 250–400 °C. The first weight loss is attributed to the loss of water (Fig. 3a–c). The native medium- and low-amylose rice starches (Fig. 3a and b) had higher initial weight loss than the acetylated starches, with values around 10% in the range of 40–125 °C, while the acetylated starches showed losses around 6% of weight in the same range. By increasing the temperature from 250 to 400 °C, the medium- and low-amylose acetylated rice starches under different times of reaction showed similar behavior, losing approximately 70% of weight. This shows that acetylation

Table 2

Thermal properties of native and acetylated high-, medium- and low-amylose rice starches.

Sample	Time of reaction	T_o (°C)	T_p (°C)	T_c (°C)	ΔH (J g ⁻¹)
High-amylose	Native	59.79	65.70	71.05	7.28
	10 min	54.90	63.07	67.49	1.17
	30 min	52.22	53.32	59.46	0.27
	90 min	49.90	50.22	54.60	0.01
Medium-amylose	Native	61.89	67.94	73.56	9.44
	10 min	52.28	54.80	60.04	0.07
	30 min	51.46	50.37	57.36	0.06
	90 min	45.15	47.66	50.18	0.04
Low-amylose	Native	60.62	67.36	75.70	13.57
	10 min	56.57	61.29	64.30	0.69
	30 min	50.62	55.99	65.54	0.42
	90 min	50.17	57.07	59.68	0.03

influenced the thermal behavior of starches; however, the intensity of acetylation did not affect the weight loss because there was no difference between the studied times of reaction.

The native and the 90 min-acetylated high-amylose rice starches showed lower loss of dry matter (5.5 and 3.0%, respectively) in the range of 40–250 °C, while the acetylated high-amylose starches after 10 and 30 min of reaction lose about 9.0% of dry matter. The lower weight loss in starch acetylated for 90 min of reaction indicates the higher stability of this material up to 250 °C. In the temperature range between 250 °C and 400 °C, the high-amylose rice starch acetylated for 90 min showed about 85.0% of dry matter loss, while the native starches and starches subjected to acetylation for 10 and 30 min of reaction showed about 70% of dry matter loss. On the other hand, for the low-amylose rice starch, the highest dry matter loss in the range of 250–400 °C was registered for the native starch and starch treated for 30 min.

Garg and Jana (2011) studied acetylated starches under different degrees of substitution and verified that acetylated starch samples were thermally more stable than native starch. The increase in thermal stability was due to the low amount of remaining hydroxyl groups in starch molecules after modification. The increase in molecular weight and covalent bonding due to the acetylation of hydroxyl groups were also responsible for the increased thermal stability.

The thermal properties measured by DSC of the high-, medium-, and low-amylose rice starches are presented in Table 2. The native starches showed higher gelatinization temperatures. There was no difference in the gelatinization temperatures of native rice starches as a function of the amylose content. Acetylation reduced the T_o , T_p and T_c values of rice starches, and it was verified a decrease in the gelatinization temperatures with an increase in the reaction time used for starch acetylation. The starch gelatinization is controlled, in part, by the amylopectin molecular structure and the granule structure. The decrease in gelatinization transition temperatures is in agreement with the early rupture of the amylopectin double helices and the melting of the crystalline lamellae in starches induced by the acetylation reaction. Luo and Shi (2012) and Singh, Chawla, and Singh (2004), acetylating the corn and potato starches, respectively, also reported a significant decrease in gelatinization temperatures after acetylation. Wotton and Bamunuarachchi (1979) suggested that the introduction of acetyl groups into polymer chains resulted in destabilization of starch granular structure, leading to a decrease in gelatinization temperatures.

When comparing the ΔH values of native low-, medium-, and high-amylose rice starches, it can be observed a high ΔH value for the low-amylose rice starch. This fact can be explained by the difference in relative crystallinity, since the crystallinity lamellae of starch granules requires higher energy for gelatinization than the amorphous lamellae. The acetylation provided low ΔH values

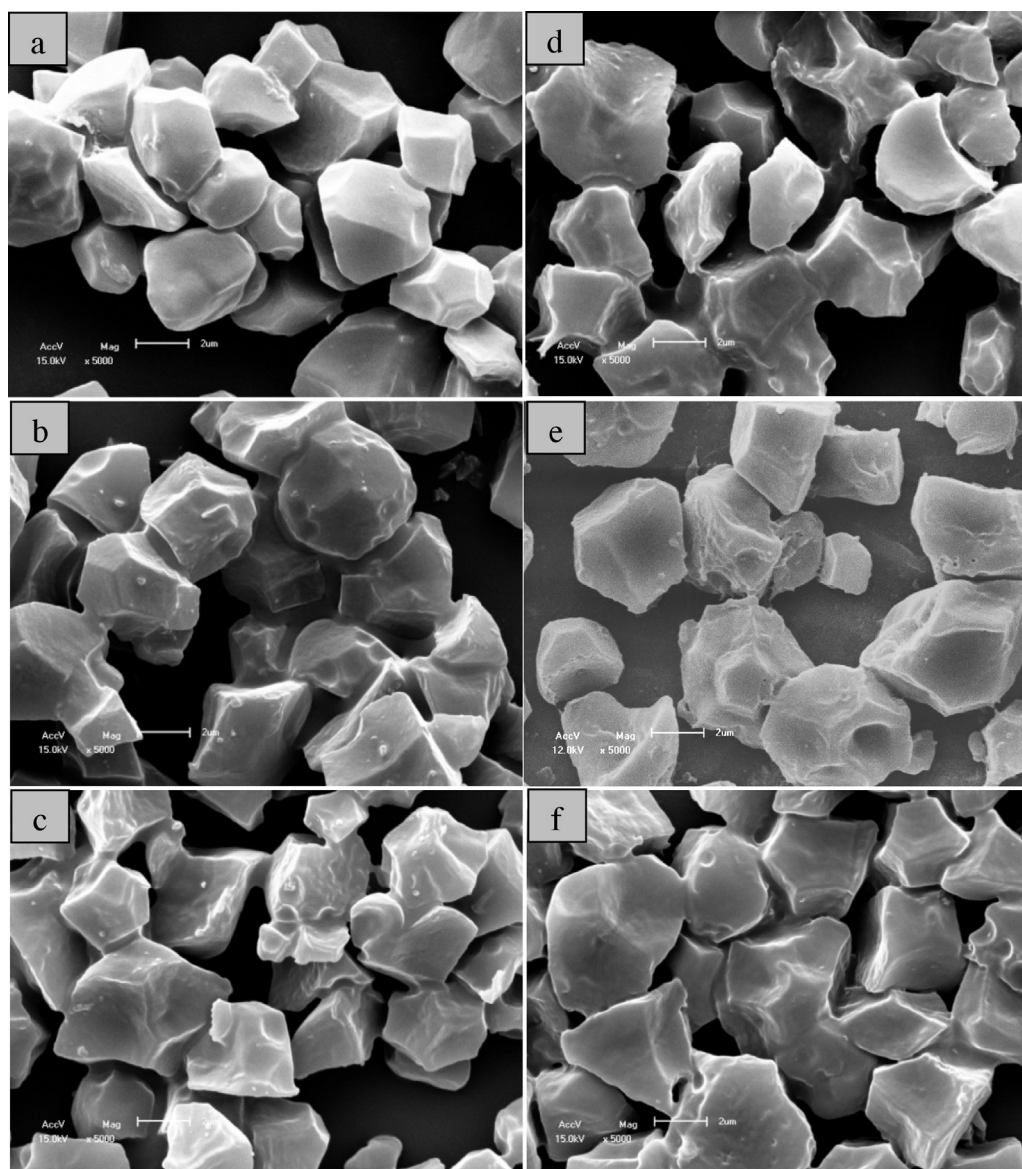


Fig. 4. Scanning electron micrographs of rice starches: native high-amylose starch (a), native medium-amylose starch (b), native low-amylose starch (c), acetylated high-amylose starch (d), acetylated medium-amylose starch (e), acetylated low-amylose starch (f). Figures d–f represent starches acetylated for 90 min of reaction.

for the high-, medium-, and low-amylose rice starches (Table 2). ΔH primarily reflects the loss of double-helical order rather than loss of crystalline register within the granule. The decrease in ΔH values of starch acetates suggests that some of the double helices present in semi-crystalline regions of the granule were disrupted during acetylation. The lower ΔH suggests a lower percentage of ordered crystallites or a lower stability of the crystals. The higher the DS of the starch, the larger the decrease in ΔH values (Table 2).

3.5. Morphology of starch granules

The morphology of starch granules was investigated using scanning electron microscopy (SEM) and the micrographs are presented in Fig. 4. The micrographs of the rice starches showed the presence of polyhedral granules. The high-, medium-, and low-amylose rice starches subjected to 90 min of acetylation (Fig. 4d–f) had higher DS and were compared with their respective native starches (Fig. 4a–c). No effect of acetylation on the morphology of starch granules was found. Sodhi and Singh (2005) also reported that the SEM revealed no significant differences between

external morphology of native and acetylated starches. However, these authors reported that the acetylation brought about slight aggregation of granules. Similar observations have been reported regarding the morphology of acetylated corn, potato (Singh et al., 2004), and rice starches (Gonzalez & Perez, 2002). Sha et al. (2012) showed that the granule surface of acetylated starch was less smooth than in native starch, but the starch granules still kept a relatively complete particle structure. As the acetyl increased, the intermolecular hydrogen bonds were damaged and more starch granules were disrupted. These authors also suggested that the crystalline regions were also involved in the reaction; the difference was that crystalline granules did not collapse.

3.6. Pasting properties

The pasting properties of native and acetylated high-, medium-, and low-amylose starches analyzed with a Rapid Visco Analyser (RVA) are shown in Table 3 and the RVA curves are presented in Fig. 5. Acetylation reduced the pasting temperature of rice starches, except for the high-amylose rice starch with the lowest DS (10 min

Table 3

Pasting properties, swelling power and solubility of native and acetylated high-, medium- and low-amylose rice starches.

Properties ^a	Time of reaction	High-amylose	Medium-amylose	Low-amylose
Pasting temperature (°C)	Native	75.42bA	70.65aB	65.85aC
	10 min	81.80aA	62.55bB	50.22bC
	30 min	61.97cA	59.55bcA	50.07bB
	90 min	nd ^b	52.77cA	53.35bA
Peak viscosity (RVU)	Native	261.66bC	290.50aB	324.37bA
	10 min	318.00aB	204.29bC	413.37aA
	30 min	285.54bB	203.54bC	393.62aA
	90 min	47.33cB	131.41cA	107.83cA
Breakdown (RVU)	Native	30.46bcA	51.08aA	178.25bB
	10 min	65.87aB	23.96bC	284.62aA
	30 min	36.83abB	25.12bB	280.92aA
	90 min	4.38cB	1.50cB	80.71cA
Final viscosity (RVU)	Native	349.54aA	347.21aA	197.08bB
	10 min	325.21abA	284.87bB	318.37aA
	30 min	312.42bAB	290.00bB	321.42aA
	90 min	104.12cB	223.62cA	55.58cC
Setback (RVU)	Native	118.33aA	107.79aA	50.96bB
	10 min	73.08bC	104.54aB	189.62aA
	30 min	63.71bC	111.58aB	208.71aA
	90 min	61.16bB	93.71aA	28.46cC
Swelling power (g/g)	Native	17.84aB	22.18aA	17.15aB
	10 min	11.27cC	13.91cB	18.75aA
	30 min	13.33bB	16.44bA	16.07aA
	90 min	7.62dB	9.82dA	9.51bA
Solubility (%)	Native	12.74aA	11.29bB	6.09aC
	10 min	11.59aA	8.46cB	4.85bC
	30 min	12.02aA	13.71aA	2.98cB
	90 min	7.27bB	11.18bA	4.26bC

^a Results are the means of three determinations. Values accompanied by lowercase letter in the same column and uppercase letters in the same row, for each property, statistically differ ($p < 0.05$).

^b nd, non-detected.

of reaction). According to Saartrat, Puttanlek, Rungsardthong, and Uttapap (2005), the pasting temperature showed lower value in acetylated starch than in native starch, and decreased as the acetyl groups content increased. This characteristic is one of the many advantages achieved with acetylation, because it allows suggesting the use the acetylated starches in processes where a thickening agent must gelatinize at lower temperatures, or simply to reduce energy costs during the manufacture of products in which these starches are used (Betancur, Chel, & Canizares, 1997).

The high-amylose rice starch acetylated for 10 min and the low-amylose rice starches acetylated for 10 and 30 min of reaction had higher peak viscosities than their native starches. When 90 min of reaction were used for the high- and low-amylose rice starches, the peak viscosity decreased compared to their native starches. The peak viscosity of medium-amylose rice starches subjected to all DS had lower values than the native medium-amylose rice starch. Acetylation reduced the final viscosity of rice starch, except for the high-amylose rice starch acetylated for 10 min and the low-amylose rice starch acetylated for 10 and 30 min of reaction, which showed equal and increased final viscosity, respectively, compared with their native starches. Saartrat et al. (2005) also found that the viscosities of acetylated canna starches were lower than those of native starches.

The marked decrease in the viscosity of the high-, medium-, and low-amylose rice starches acetylated for 90 min (Fig. 5) cannot be attributed to the partial gelatinization of starch granules, since there was no loss of granular integrity according to the SEM (Fig. 4d–f). The decrease in the viscosity of acetylated starches compared to native starches can be attributed to the insertion of acetyl groups that hinder the association between starch chains and decreased the ability of starch granules to absorb water. Thus, it gives starch a hydrophobic character.

Acetylation reduced the breakdown of rice starches, increasing the thermal and mechanical stability of acetylated starches, except for the high- and low-amylose rice starches acetylated for 10 and 30 min of reaction. The high-amylose rice starches acetylated at all DS and the low-amylose rice starch acetylated for 90 min of reaction had a lower setback compared to their native starches. There was an increase in the setback of low-amylose rice starch acetylated for 10 and 30 min compared to the native low-amylose rice starch. The reduction in the setback is due to the introduction of acetyl groups in starch chains, which can prevent close parallel alignment of amylose chains and thus lower setback viscosities. However Sodhi and Singh (2005) found that acetylated starches show higher setback viscosities than their native counterparts. Such effect was observed for medium-amylose starches and for low-amylose starch acetylated for 10 and 30 min of reaction (Table 3).

3.7. Swelling power and solubility

Acetylation reduced the swelling power of rice starches, except for the low-amylose rice starch when acetylated for 10 and 30 min, which showed swelling power similar to native starch. The highest decrease in swelling power was verified in starches acetylated for 90 min of reaction (Table 3), which exhibited high DS (Table 1). In starches modified by acetylation, the introduction of hydrophobic acetyl groups can make the water intake into starch granules difficult, thus decreasing the swelling power.

Comparing the rice starches with different amylose contents, the low-amylose starch showed the lowest solubility compared to the high- and medium-amylose rice starches, which is probably due to the lower amount of amylose molecules that are leached during hydration and heating. The decrease in starch solubility is due to the lower amylose leaching and can be a result of the higher interaction

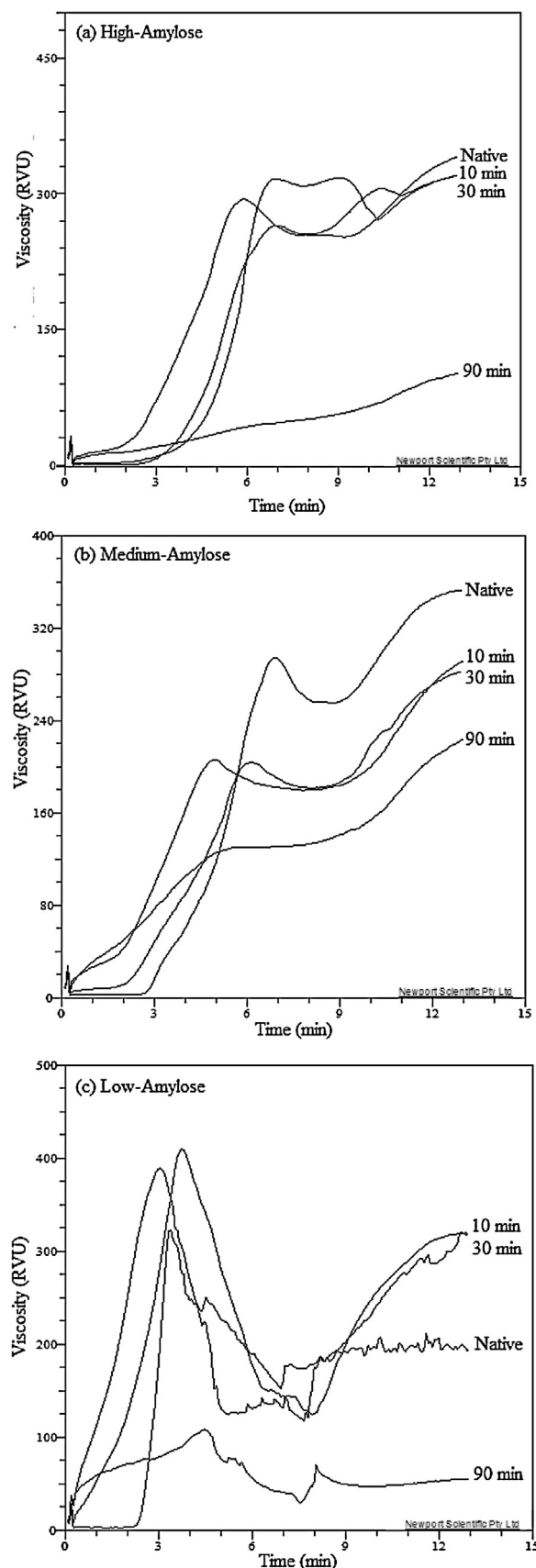


Fig. 5. RVA curves of native and acetylated rice starches. High-amylose starch (a), medium-amylose starch (b), and low-amylose starch (c).

between amylose and amylopectin molecules, preventing the amylose from leaching from the granule. The increase in the molecular weight of starch due to the introduction of acetyl groups mainly in C(6) may make the leaching of amylose from the starch granule difficult. The solubility characteristic of the acetylated starch is dependent of the DS and the polymerization of amylose and amylopectin chains. Lawal (2004) also found similar trends of decreased solubility from new cocoyam starch acetylated with 60 min of reaction when compared with native starch.

4. Conclusions

The present study was the first one about acetylation of rice starch of different amylose contents. The low-amylose rice starch was more susceptible to acetylation compared to the medium- and high-amylose rice starches. The introduction of acetyl groups was confirmed by FTIR spectroscopy. Acetylation, mainly over 90 min of reaction, reduced rice starch crystallinity and, in general, its pasting temperature, breakdown, peak and final viscosities, swelling power, and solubility. The decrease in pasting temperature and breakdown of rice starches enables obtaining products sensitive to high temperatures and more stable products while cooking. The continuity of this work should evaluate the susceptibility of acetylated starches with different amylose content and DS to enzymatic hydrolysis, as well as the production of biodegradable films using acetylated rice starch.

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