

Thermal and physicochemical properties of starches from three Colombian rice varieties

Propiedades térmicas y fisicoquímicas de almidones de tres variedades de arroz colombianas

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ABSTRACT

Samples of starch from broken grains of three rice varieties grown in Colombia were analyzed to determine their physicochemical and thermal properties: Fedearroz 473 (F473), Fedearroz 50 (F50) and Fedearroz 60 (F60). The granule size, solubility, swelling capacity, amylose content, syneresis, turbidity, thermal and pasting properties of starches were determined. The average size of starch granules was 9.4, 7.4 and 7.2 μm for F473, F50 and F60 samples, respectively. The amylose content showed significant differences between the studied varieties and ranged between 21.4% and 23.0%. Turbidity ranged between 1.95 and 2.34 absorbance units at 620 nm. Thermal properties, evaluated by differential scanning calorimetry (DSC), registered values between 61.6 and 64.6°C for the onset temperature, between 66.6 and 69.3°C for the peak temperature, between 72.1 and 73.9°C for the end temperature, and between 8.38 and 9.47 J g^{-1} for the gelatinization enthalpy. The higher amylose content the higher grain size, turbidity, syneresis, viscosity, gelatinization temperature and enthalpy, and the lower swelling capacity and solubility. This paper is the first reported research on physicochemical and functional properties of starches from these Colombian rice varieties.

Key words: *Oryza sativa*, swelling capacity, gelatinization, solubility, amylose content.

RESUMEN

Muestras de almidón obtenidas de arroz partido de tres variedades cultivadas en Colombia fueron analizadas para determinar sus propiedades fisicoquímicas y térmicas: Fedearroz 473 (F473), Fedearroz 50 (F50) y Fedearroz 60 (F60). Se determinó el tamaño de gránulo, la solubilidad, el poder de hinchamiento, el contenido de amilosa, la sinéresis, la turbidez, las propiedades térmicas y las propiedades de empastamiento de los almidones. El tamaño granular promedio de las muestras F473, F50 y F60 fue de 9,4; 7,4 y 7,2 μm respectivamente. El contenido de amilosa mostró diferencias significativas entre las variedades estudiadas y sus valores oscilaron entre de 21,4 y 23,0%. La turbidez osciló entre 1,95 y 2,34 unidades de absorbancia a 620 nm. Las propiedades térmicas, evaluadas mediante calorimetría diferencial de barrido (DSC), registraron valores entre 61,6 y 64,6°C para la temperatura de inicio, entre 66,6 and 69,3°C para la temperatura pico, entre 72,1 y 73,9°C para la temperatura de finalización, y entre 8,38 y 9,47 J g^{-1} para la entalpía de gelatinización. Los almidones con mayor contenido de amilosa mostraron un mayor tamaño granular, turbidez, sinéresis, temperatura y entalpía de gelatinización, viscosidad, y un menor poder de hinchamiento y solubilidad. El presente estudio es la primera investigación reportada en propiedades fisicoquímicas y funcionales de almidones de arroz provenientes de estas variedades cultivadas en Colombia.

Palabras clave: *Oryza sativa*, capacidad de hinchamiento, gelatinización, solubilidad, contenido de amilosa.

Introduction

Rice is one of the main cereal crops worldwide and its starch in one of the key ingredients of several food products. According to commercial importance, there are over two thousand rice varieties grown all around the world (Deepa *et al.*, 2008).

FAOSTAT (2017) informed that rice production in 2013 was around 732 Mt, from which, around 23 Mt were produced in Latin America and the Caribbean. Colombia has an outstanding position in Latin America due to the varieties

provided by the National Federation of Rice (Fedearroz). Over 30 varieties have been released since 1970. From 1997, F50, Colombia XXI, F2000, F473, F369, F275, and more recently, F60 and F174 varieties have been released.

In the first semester of 2016, Colombia achieved a rice production of 765,355 t, from which the province of Tolima produced approximately 38% (Dane, 2017). Tolima has the most competitive milling industry of the country, thanks to its degree of entrepreneurial development, supply lines, infrastructure and closeness to the most important urban

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markets. However, not all rice production is commercialized as whole or polish grains. The broken or small grains could be lower than a quarter of a grain in size and they are often used in animal feeding and beer industry.

According to Fedearroz (Colombia), there is a strong difference in rice prices according to its quality, which leads to the need of setting other uses that generate added value that can be more representative in economy and thus, increase the yield of rice production chain, additionally associated to the innovation process which grants an alternative to the industry for exploitation and optimization of the resources facing global markets.

If it is considered that FAO (2006) observes a series of opportunities in the production of starch that should be taken advantage of, in the Tolima region there are important limitations faced by the industry pointed out, such as irregularity in the supply and the unequal quality of the final product. Just the same, there is a dissatisfied demand of starch, which creates the necessity of finding new sources of such product.

The goal of the current study is to assess physicochemical and thermal properties of native starches from rice varieties grown in Tolima province and determine their functionality concerning their assessed properties.

Materials and methods

Starch extraction

Starch extraction from varieties Fedearroz 473 (F473), Fedearroz 50 (F50) and Fedearroz 60 (F60) was carried out based on the method by Devi *et al.* (2009). The samples were supplied by Las Lagunas Experimental Center of Fedearroz in Saldaña (Colombia).

Amylose content

The determination of amylose was carried out by Iodine-based colorimetric method described by Juliano (1985). Iodine-based colorimetric method remains the most widespread method for the determination of amylose content of starch (Mahmood *et al.*, 2007). Potato amylose (analytical grade) was used as reference.

Microscopic appearance

Shape and diameter of starch granules were determined using a fluorescence microscope (Axio Imager A1, Carl Zeiss, Germany) and the provided software (Axio Vision AC rel. 1, Carl Zeiss, Germany).

Swelling capacity and solubility

Starch suspensions (2%, w/v) were preheated in a water bath at 90°C for 30 min and were centrifuged at 2,000 rpm for

20 min. The supernatants were withdrawn and sediments were weighed. Supernatant aliquots were dried at 100°C in a convective oven until reaching a constant weight. Swelling capacity (g g^{-1} , dry basis) and solubility (%) were calculated as equation (1) and (2), respectively.

$$\text{Swelling capacity} = \frac{\text{Sediment mass}}{\text{Dry starch mass} \times (100 - \text{dry supernatant percentage})} \quad (1)$$

$$\text{Solubility} = 100 \times \frac{\text{Dry supernatant mass}}{\text{Dry starch mass}} \quad (2)$$

Turbidity

Turbidity was measured according to Craig *et al.* (1989). An aqueous suspension of starch at 2% was heated in a boiling water bath for 1 h with constant stirring. The suspension was cooled down for 1 h at 30°C and turbidity was determined measuring absorbance at 620 nm. Distilled water was used as blank.

Syneresis

Syneresis was measured according to Singh *et al.* (2006) with some modifications. A starch suspension (5% w/v, dry starch mass) was heated in a water bath at 90°C for 20 min. After cooling at ~25°C, samples were located in a freezer at -10°C during 48 h and then put in a water bath at 30°C to defreeze until thermal equilibrium. Syneresis was measured in percentage as the quantity of water released after centrifuging at 2,500 rpm for 40 min.

Thermal properties

Differential scanning calorimetry (DSC) experiments were performed using a Perkin Elmer DSC 8000 (Perkin Elmer, Waltham, MA, USA). The instrument was calibrated with indium (melting point 156.6°C, fusion enthalpy 28.45 J g^{-1}) at a heat flux of 5°C/min. Aluminum pans (ref 0219-0041, Perkin Elmer, Waltham, MA, USA) were used as recipients for the samples. For sample preparation, approximately 2 mg of starch were put in aluminum capsules for DSC analysis and distilled water was added (1:2 w/w starch:water) (Vandeputte and Delcour, 2004). Capsules were weighed, sealed and stored at room temperature for 24 h. DSC experiments were carried out under nitrogen atmosphere (99.5% purity, ~20 mL min^{-1} gas flow) and at a heating rate of 5°C/min from 30°C to 110°C. The PYRIS 10.1 software (Perkin Elmer, Waltham, MA, USA) was used to analyze the thermal data. The gelatinization onset, peak and end temperatures, and gelatinization enthalpies were determined in order to characterize the thermal behavior of rice starches. Results were averages of eight measurements for each rice variety.

Pasting properties

Each starch sample (at ~10% moisture content) was suspended in distilled water to obtain 10% suspension. The amylograms were obtained using a micro Brabender Viscoamylograph (Brabender OHG Duisburg, Germany) where suspensions were heated from 30 to 95°C at 10°C/min, held at this temperature for 3 min, and cooled to 30°C at 10°C/min (Suh and Jane, 2003).

Statistical analysis

Statistically differences between the three rice varieties were assessed via one-way analysis of variance (ANOVA), at a significance level of 95%, for granule size, solubility, swelling capacity, syneresis, amylose content, turbidity and thermal properties. Multiple range tests (MRT) were carried out to determine which means are significantly

different from which others using the Fisher's least significant difference (LSD) procedure. ANOVA and MRT were performed using Statgraphics Centurion XV software (StatPoint Technologies Inc., USA).

Results and Discussion

Amylose content

F50 and F60 varieties did not show significant differences for amylose content, whose values were 21.4 and 21.5% respectively, as shown in figure 1A. Starch from variety F473 showed significant variations concerning other varieties and the highest content of amylose (23.0%). Data collected for amylose content are similar to the ones reported by Patindol *et al.* (2007) (between 17.0 and 21.6%) and by Falade and Christopher (2015) (between 20.7 and 26.0%).

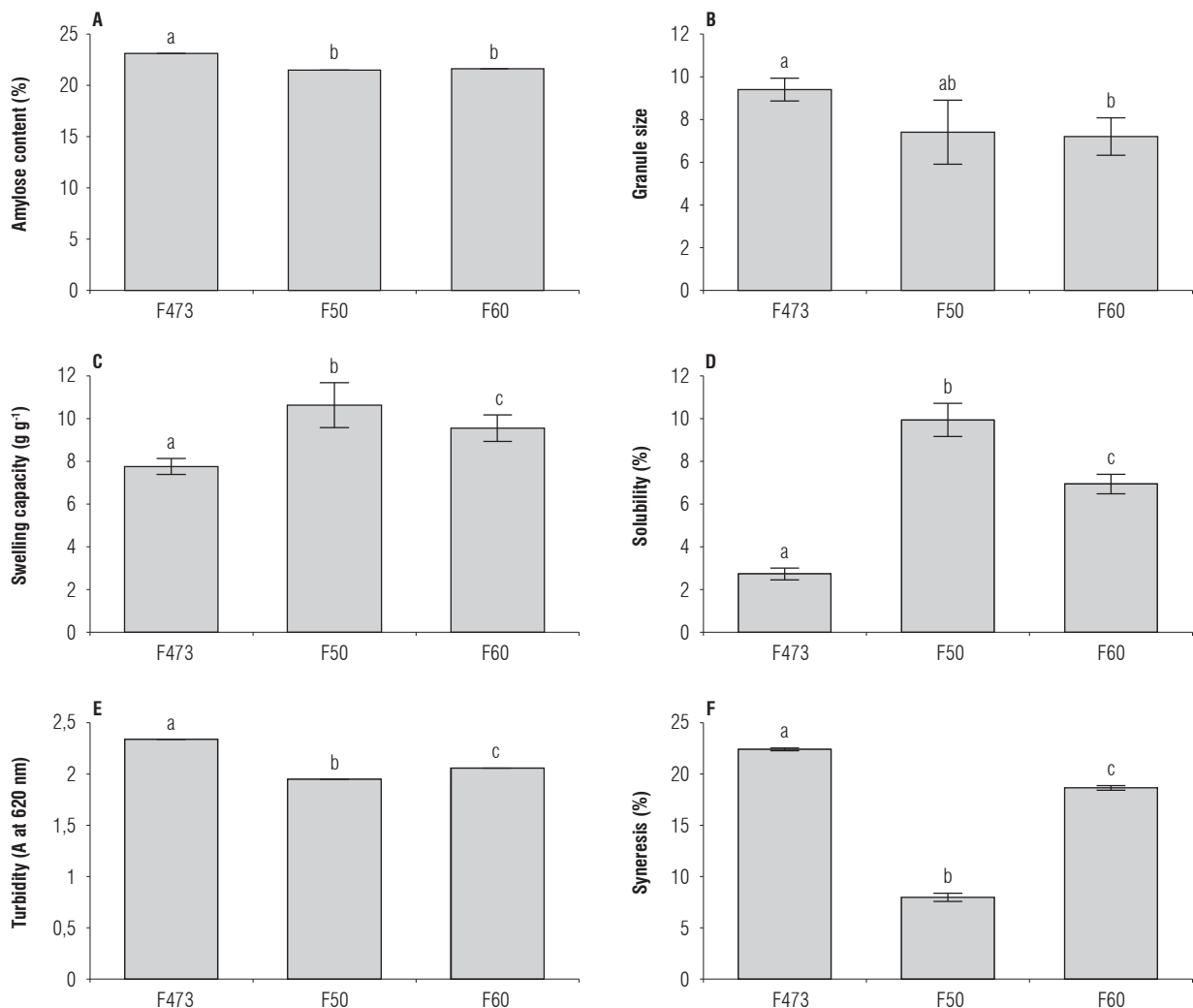


FIGURE 1. Amylose content (A), granule size (B), swelling capacity (C), solubility (D), turbidity (E) and syneresis (F) of starches from F473, F50 and F60 rice varieties. Error bars represent standard deviation from the mean. Means with different letters indicate significant difference according to LSD test ($P \leq 0.05$).

It has been reported that differences in amylose content of starch vary due to weather and agronomic conditions during grain development (Singh *et al.*, 2006; Wang *et al.*, 2002; Wang *et al.*, 2010).

Amylose content seems to be the main factor that controls many of the physicochemical properties of rice starch, such as turbidity, syneresis, pasting, gelatinization and retrogradation properties (Wickramasinghe and Noda, 2008). Just the same, it plays a key role in starch digestion. The ones that show low amylose content are more easily digested than the ones with a higher content (Riley *et al.*, 2004).

Microscopic appearance

Microscopic appearance of starch granules is observed in figure 2. Analyzed starches showed polyhedral and irregular shapes as reported by Sodhi and Singh (2003). Average granular size of F473, F50 and F60 varieties was $9.43 \pm 0.43 \mu\text{m}$, $7.43 \pm 1.50 \mu\text{m}$ and $7.23 \pm 0.88 \mu\text{m}$, respectively; similar data to the ones reported by Yang *et al.* (2006). Starches from varieties F50 and F60 did not show significant differences; on the other hand, starches from varieties F473 and F60 exhibited significant variations, as shown in Figure 1B.

Singh *et al.* (2003) concluded that amylose content varies with granule size; the higher average size the higher amylose content. Starch from variety F473, which presents the highest average diameter, also shows the highest amylose content. The other two starches (F50 and F60) whose average granule sizes are lower than the one from F473 variety, but without significant difference between them, presented lower amylose contents.

Differences shown between size and structure of starch granules match their botanical origin (Hoover, 2001). Rice starch granules are the smallest among cereal grains, with a size between 2 and $7 \mu\text{m}$ (Vandeputte and Delcour, 2004). Variation, particularly in size and granule form is associated to different functional properties in diverse alimentary systems, such as pasting and mixing, as well as to the possibility of relating granule morphology to elaboration processes or nutritional qualities (Peterson and Fulcher, 2001).

Swelling capacity and solubility

Swelling capacity and water absorption occurs due to water adherence to the surface of starch granules and leads the granules to swell. This behavior can be attributed to the relation amylose/amylopectin, given that water particles are trapped in amylopectin structure, as well as the difference in the distribution of the chain length (Bello-Perez *et*

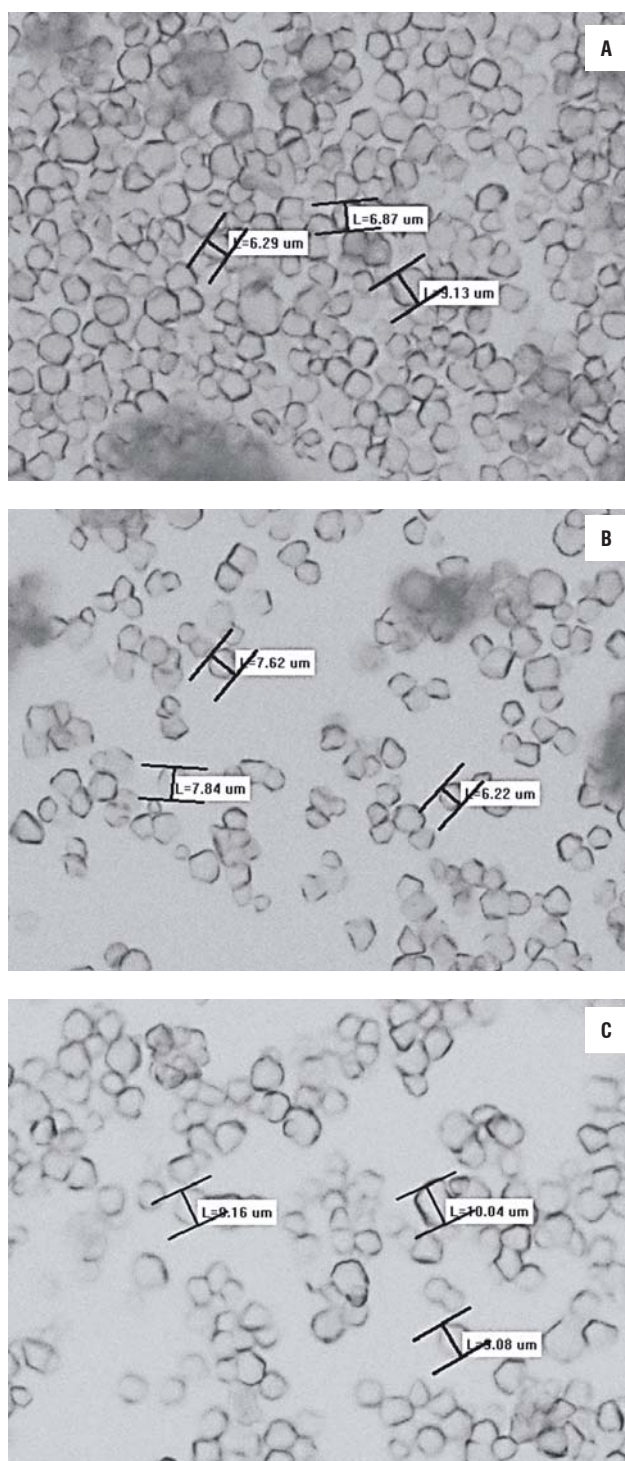


FIGURE 2. Microphotographs of rice starch granules. F50 (A), F60 (B) and F473 (C).

al., 1998). Just the same, Cai *et al.* (2015) determined that swelling capacity is positively related to short amylopectin chains and their branching degree. In the same way, a positive relation between solubility and amylose content was determined.

Values of swelling capacity for F473, F50 and F60 varieties showed significant differences, as shown in Figure 1C. Swelling capacity values are similar to the ones reported by Wickramasinghe and Noda (2008); values ranging between 7.33 and 16.12 g g⁻¹. The behavior of swelling capacity concerning to amylose content was coherent with those reported by Sodhi and Singh (2003) and Wang *et al.* (2010), who concluded that there is a negative correlation of these two properties.

Starch from variety F50, whose amylose percentage was the lowest, showed the highest swelling capacity (10.67±1.06 g g⁻¹). In the same way, starch from variety F473, whose percentage was the highest, showed the lowest swelling capacity (7.79±0.38 g g⁻¹).

Concerning solubility, starches from varieties F473, F50 and F60 presented significant differences with values of 2.71±0.27%, 9.85±0.27% and 6.88±0.45%, respectively (Fig. 1D). Solubility values obtained by analyzed starch are similar to the ones reported by Chang *et al.* (2010), which obtained solubility values for native starches under 10% (8.5, 7.5 and 7.4%). Lin *et al.* (2011) reported that there is a negative correlation between amylose content and solubility. Starch of F473 variety, whose amylose content was the highest, presented the lowest solubility value (2.71 ±0.27%). Just the same, F50 variety starch showed the highest solubility (9.85±0.77%).

Although the F50 and F60 varieties had similar amylose contents, they showed significant differences between swelling and solubility that could be attributed to other factors not considered in this study, such as the branching degree of the amylopectin (Wang *et al.*, 2010).

Turbidity

Turbidity is inversely related to the transparency of pastes, the dispersion of the solutes and the tendency to the retrogradation of the starches. The capacity to transmit light when exposed to a ray of light passing through these pastes defines its clarity. Turbidity values showed significant differences among varieties (Fig. 1E). Starches that show a higher quantity of amylose are more difficultly dispersed, which increase their turbidity, just like the ones that show a higher swelling capacity (Novelo and Betancur, 2005). This behavior occurred in F50 variety starch, showing higher clarity, lower amylose content (21.28%) and a higher swelling capacity (10.66 g g⁻¹). However, absorbance values registered for analyzed samples are higher than the ones reported for rice starch, which allows for assumption that pastes are more opaque and make them feasible for food clouding.

Syneresis

Syneresis values of analyzed starches showed significant differences. Values ranged between 8.0 and 22.3%, as shown in figure 1F. The starch that presented the best stability to freezing–defrosting was F50. The highest syneresis was given by F473 starch (22.3%) since it presented low stability in freezing–defrosting processes, causing loss of water trapped in gel. Some researchers have reported low syneresis values (between 0.01 and 1.81%) for rice starch stored at 4°C for 24 h (Singh *et al.*, 2003; Singh *et al.*, 2006). Syneresis values obtained in analyzed starches are higher than the ones reported by Sodhi and Singh (2003), whose records were located between 0.04 and 2.41% in 48 h. On the same way, analyzed starches registered lower values than the ones given by Wang *et al.* (2010), whose syneresis was between 22.9% and 46.4% in 22 h.

Starch from variety F473, which presented highest syneresis and amylose content, showed the lowest swelling capacity and highest turbidity. This behavior is coherent with that reported by Singh *et al.* (2006), who concluded that there is a negative correlation between swelling capacity and syneresis, and a positive correlation between the latter one and turbidity. Such things occur because syneresis happens due to the increase of molecular association among starch chains, at reduced temperature, thus eliminating water from the gel structure. For gels of waxy rice starch, or with little amylose, there is a higher resistance to syneresis, because of the formation of a lower number of intermolecular associations (Bao *et al.*, 2004).

Sodhi and Singh (2003) established that there is a positive relationship between granule size, amylose content, syneresis and turbidity. Such behavior corresponds to the values of analyzed starches, from which, starch from F473 variety presented the highest granule size, the highest amylose content and the highest syneresis percentage. Despite the relationship of such properties with amylose content, F60 and F50 showed statistically significant differences between syneresis and turbidity, which could be attributed to differences in the degree of branching of amylopectin chains, as mentioned above.

Thermal properties

Gelatinization onset (T_o), peak (T_p) and end (T_e) temperatures for F473, F50 and F60 rice varieties are shown in Figure 3. Gelatinization enthalpies (ΔH) were 9.47±0.38 J g⁻¹, 8.62±0.33 J g⁻¹ and 8.38±0.34 J g⁻¹ for F473, F50 and F60, respectively. Varieties F50 and F60 did not show significant differences for T_o and ΔH . The highest T_o was registered from variety F473 (64.6°C). Average T_o values for

were similar to the ones reported by Noosuk *et al.* (2003), who determined onset temperatures around 62.7°C for Thai rice starches.

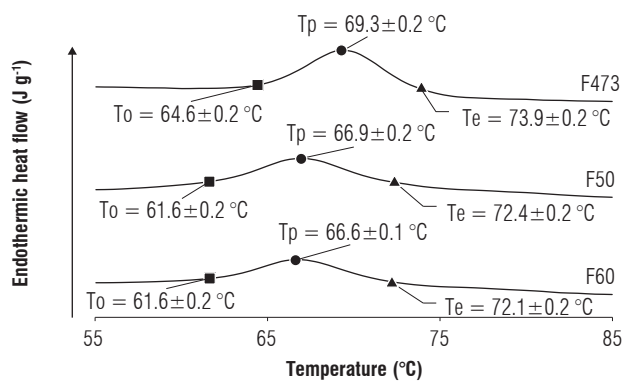


FIGURE 3. Thermal properties of starches from F473, F50 and F60 rice varieties. Onset temperature (T_o), peak temperature (T_p) and end temperature (T_e).

Analyzed starches showed significant differences for T_p and T_e . F473 presented the highest T_p with a 69.31°C. For T_e , starches from the three varieties showed significant differences, being F473 the one with the highest value (73.90°C). The average values presented by analyzed starches were similar to the ones reported by Wang and Wang (2004) (73.1°C) for starch from long-grain rice flour.

Gelatinization enthalpy (ΔH) reflects the loss of molecular order (Cooke and Gidley, 1992). Values of ΔH of analyzed starches are similar to the ones reported by (Singh *et al.* 2007), whose values were between 8.2 and 9.8 J g⁻¹ for different *indica* rice cultivars.

According Wang *et al.* (2010), there is a positive correlation between ΔH and gelatinization temperatures. Variation in T_o and ΔH in starches of different varieties could be due to

differences in the quantities of longer amylopectin chains. These longer chains require a higher temperature to dissociate completely than the required for short double helices (Yamin *et al.*, 1999). This correlation was evidenced in F473 variety starch, whose enthalpy was the highest with a value of 9.47 J g⁻¹, just like the highest T_o , T_p and T_e .

Concerning the relation between amylose content and gelatinization temperatures, contradictory studies have been found. Szczodrak and Pomeranz (1992) concluded that lower enthalpy values are related to higher amylose contents. Sodhi and Singh (2003) concluded that starches with a lower polysaccharide content presented a higher swelling capacity and transition temperatures. Varavinit *et al.* (2003) informed that there is a positive correlation of gelatinization with amylose content; this behavior was registered from starches of F473, F50 and F60 varieties. On the other hand, some studies (Singh *et al.*, 2006; Park *et al.*, 2007) did not report correlations between amylose content and thermal properties.

Pasting properties

Just as shown in table 1, temperatures at the beginning of gelatinization by viscosography for F50, F60 and F473 varieties were 67.7, 67.2 and 70.8°C respectively; similar data to the ones reported previously by DSC, which confirm the behavior of analyzed varieties.

In figure 4, viscosity behavior is observed concerning time and temperature of F473 starch, which presented the maximum viscosity with 196 Brabender units (BU) and the highest amylose content. This behavior is coherent with the information reported by Singh *et al.* (2006), who established a positive correlation between these two properties. The amylogram shows a tendency to retrogradation, producing the syneresis process. The presence of amylose favors retrogradation during cooling period

TABLE 1. Pasting properties of starches from F473, F50 and F60 rice varieties.

Assessment point	Time (min)			Viscosity (BU)			Temperature (°C)		
	F50	F60	F473	F50	F60	F473	F50	F60	F473
Gelatinization start	6.2	5.97	6.3	13	12	11	67.7	67.2	70.8
Maximum viscosity	8.8	8.8	10.2	182	168	196	82.1	82.1	88.5
Start of maintenance period	10.8	10.8	10.8	146	143	186	93.7	93.7	92.3
Start of cooling period	13.8	13.8	13.8	136	124	161	95.4	95.3	94.5
End of cooling period	21.3	21.3	21.3	257	251	276	64.5	64.4	65.1
End of maintenance period	23.3	23.3	23.3	295	292	285	58.7	55.0	57.4
(Stability) Breakdown	0.0	0.0	0.0	46	44	35	0.0	0.0	0.0
(Final) Setback	0.0	0.0	0.0	120	127	115	0.0	0.0	0.0

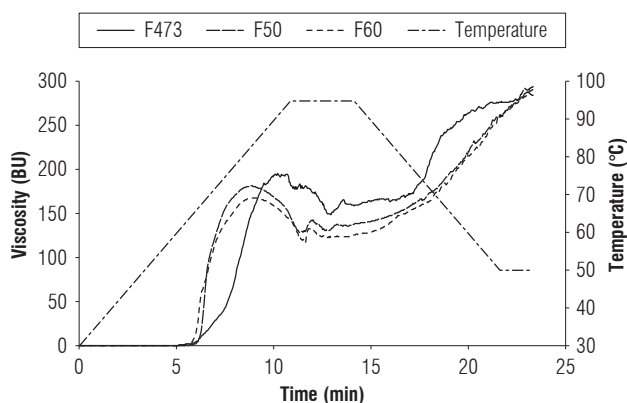


FIGURE 4. Amylograms of starches from F473, F50 and F60 rice varieties.

due to the annealing of the soluble starch polymers and the insoluble granular fragments (Hoover, 2002). Besides, this starch presented the highest syneresis value, highest gelatinization temperatures, higher paste stability and the lowest cooking easiness. The paste stability is related to the variation of the maximum viscosity and the viscosity during the maintenance period, while the cooking easiness corresponds to the difference between the time to reach the maximum viscosity and the gelatinization start.

For the case of F50 starch, figure 4 shows a maximum viscosity of 182 BU, the highest gel instability with a value of 46 BU and greater ease of cooking with 2.6 min. This starch presented the lowest amylose content and showed tendency to retrogradation. The difference of behaviors in the cooking easiness can be attributed to the fact that there is presence of starch granules of F50 variety that occupy a bigger surface area in the solution, meanwhile starch granules of F473 variety, with a bigger diameter, has a higher incidence in cooking time (Hoover, 2002).

For the case of F60 starch, the Figure 4 shows a maximum viscosity of 168 BU, gel instability with a value of 44 BU and a cooking easiness of 2.8 min in the amylogram, a tendency to retrogradation can be observed.

Differences in viscosity are mostly due to amylopectin presence in starch (Lin *et al.* 2011), since it is the polymer that solubilizes fastest in aqueous means and gives viscoelastic stability to the pasting curve, when this solution is exposed to sudden temperature changes (Wang *et al.*, 2003)

Food applications

Starches with a higher amylose content presented bigger granule size, higher turbidity, maximum viscosity and temperature, and gelatinization enthalpy. On the contrary,

they showed the lowest values in swelling capacity and solubility. In food applications, it is desirable that starches have a high swelling capacity and viscosity as well as low solubility and syneresis. However, from the functional point of view, properties limit the specific use of starches in the different applications (Granados *et al.*, 2014).

Swelling capacity and solubility of native rice starches are lower than most starches from other sources used in the industry. This behavior can be associated to the size granule that, in the case of rice starches, is smaller than from other sources and to amylose content, generally presented in a higher proportion. Low swelling capacity of rice starch restricts it as humidity retainer.

Syneresis in rice starches presented a relatively lower value than starches from other botanical sources, possibly due to the fact that starches in other sources have different retrogradation rates and at different degrees. Just the same way, amylose content and amylopectin chains length determine this phenomenon. Syneresis value presented by starches makes them feasible in the use of products that require a certain exudation degree within their appearance.

From the point of view of functional behavior, starch from studied varieties presents low solubility, low syneresis and a small size of the granule (better palatability). On the same way, opaque pastes that make them feasible to be used as clouding agents

Physicochemical and thermal properties of starches from three rice varieties grown in Colombia were evaluated in order to establish potential uses. The study determined that low swelling capacity of starches in varieties F50, F60 and F473, does not make them eligible to be used as humidity retainers (e.g. meat products). Otherwise, by showing high absorbance values and presenting opaque pastes, they are feasible to be used in slightly transparent products, such as mayonnaises, concentrated drinks or bakery products. Just the same, the low syneresis presented by F50 variety enables it to be used in soups, cake fillings and child foods. On the contrary, F60 and F473 varieties are potential for the use of foods that require certain exudation level like sauces or dressings. Low gelatinization temperatures do not make them desirable in products that require high processing temperatures such as canned foods. Instead, they can be used in the elaboration of custards and puddings (Hernández *et al.* 2008). Starch from variety F473 showed the best gel stability, which makes it eligible for foods that require cooling in their process.

Conclusions

The present study is established as the first analysis carried out on rice starches of Colombian varieties recorded so far. Starch from F473 variety showed significant differences with F50 and F60 varieties regarding to amylose content, syneresis, swelling, solubility, turbidity and thermal properties. These differences are largely due to each variety as well as amylose content and granule size. On the other hand, the analyzed starches could be applied in the food sector, especially as clouding, but not as a moisture retainer.

Additional studies should be carried out with the analyzed varieties in order to evaluate a greater number of properties, and thus establish with a wider range of industrial uses.

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