Physical, chemical and rheological properties of pitomba (Talisia esculenta) seed starch and its application as a thickener and stabilizer in ketchup

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Abstract

The present study aimed to characterize the physical, chemical and rheological properties of starch extracted from pitomba seeds and verify the influence of its addition on the syneresis and viscosity characteristics of ketchup during storage, compared to maize starch. Extraction was performed by grinding the seeds in sodium metabisulfite solution (0.2%) followed by drying at 50 ºC. Ketchups were formulated with the addition of 0.5 and 1% pitomba starch, 0.5 and 1% maize starch and a control formulation. Starch showed low percentages of protein, ashes and lipid, starch purity of 77.78%, amyllose of 19.25%, apparent density of 0.46 g/cm³ and compact density of 0.66 g/cm³. The starch pastes showed pseudoplastic behavior and imparted significant viscosity, usable in increasing the consistency of fluid foods. Ketchups formulated with addition of pitomba starch showed reduction of syneresis during storage, and the 1% starch concentration can slow down the syneresis process in ketchup at temperatures of 7 and 25 ºC, similar to the stabilizing capacity of maize starch. Ketchup viscosity increased with the addition of pitomba and maize starches, but the highest apparent viscosities were obtained in ketchup with addition of pitomba starch during storage.

Keywords: consistency; flowability; native starch; rheological properties; syneresis.

Introduction

Pitomba (Talisia esculenta) is a fruit native to the Western Amazon and its cultivation is carried out in an unorganized manner, in the North and Northeast regions of Brazil. Pitomba is only consumed fresh because the difficulties to remove the pulp and low yield hamper the processing technologies, but its endocarp represents about 50% of the total fruit weight, constituting a source of starch (Neri-Numa et al., 2014; Castro et al., 2019).

Starch has been traditionally used to maintain the quality of processed foods, acting as ingredient or additive to improve their properties, serving as thickening and gelling agent, controlling water retention, and stabilizing the texture of processed foods (Zhang et al., 2016). Starch functionally is related to its physical-chemical variations, which are influenced by the molecular structure, plant sources from which it is extracted, gelatinization temperature, gel formation and absence or presence of chemical modification, which characterizes it as either native or modified (Rondán-Sanabria and Finardi-Filho, 2009; Correia et al., 2012; Rani and Chawhaan, 2012; Barbi et al., 2018). Native starches have limitations in industrial applications due to their high thermal instability and tendency to retrograde during cooling and freezing, but the trend of using natural and clean label ingredients has influenced the use of native starches (Arocas et al., 2009).

Sauces, such as tomato, pepper, white or rose, used to enhance the taste of foods, are frequently consumed during meals and need to meet requirements of viscosity and stability for good acceptance. The main problem associated with sauce quality is the tendency of separation of solids from the liquid phase during storage, so the food industry adds thickening and
stabilizing ingredients in these formulations and commonly resorts to the use of starches. The present study aimed to determine the physical, chemical and rheological properties of native starch extracted from pitomba seeds and verify the influence of its addition, compared to maize starch, on the viscosity and stability characteristics of ketchup during storage.

Results and discussion

Yield, moisture, crude protein, ashes, lipids, total starch and amylose
The results of yield, moisture, and physical-chemical characterization of pitomba starch are presented in Table 1. The average starch extraction yield was 44.20%, which is close to the results reported by Santhitha and Sasikumar (2015), who observed variation from 42.24 to 48.48% for starches of different varieties of curcuma. A lower yield, 30.33%, was found by Wani et al. (2016) in starch from fava beans. A moisture percentage of 8.41% was found in pitomba starch, which fits the specifications recommended by the Brazilian Legislation for commercial starches (maximum moisture content of 14%) (Brasil, 1987). The low percentages of protein, ashes and lipids reflect a good degree of purity in the sample, a desirable characteristic because the presence of these contaminants interferes with starch functional properties. Ascheri et al. (2010) recommend that lipid percentage in the starch should be lower than 1%. Values close to the one found in pitomba starch were reported by Gani et al. (2017), who characterized starch extracted from Himalayan rice cultivars and observed variation from 0.39 to 0.56%, and by Falade et al. (2014), who characterized starch extracted from African rice and observed lipid percentage of 0.40%.

The starch percentage found (77.78%) is similar to the one reported by Melo-Neto et al. (2015), for starch extracted from pejibaye fruits (79.00%), and higher than the value found for starch from Tommy Atkins mango seed (71.56%), reported by Mendes et al. (2015). The starch showed 19.25% of amylose. In the literature, a wide variation of amylose has been reported for different starches, which may be attributed to botanical characteristics and determination methods. However, values close to those found here were reported by Lemos et al. (2017), who obtained amylose percentages of 19.70, 20.46, 16.36 and 11.19 for native starches of corn, potato, banana and cassava, respectively. Amylose proportion has influence on the viscosity of starch pastes and, according to Bello-Perez et al. (2006), the low content of amylose plays an important role in starch physical-chemical and functional properties, because it is the main responsible for the retrogradation phenomenon.

Starch flow properties
The results for the flow properties of pitomba starch are presented in Table 2. Pitomba starch had low apparent density (0.46 g/cm³), which is associated with smaller particle size, causing greater porosity during powder rearrangement. The apparent density of pitomba starch is similar to the value of 0.46 g/cm³ reported by Pachaua et al. (2017), who analyzed the physical properties of starch from a variety of glutinous rice from India.

The low value obtained for compact density reveals the tendency of this starch to become compacted during storage, which can alter the handling properties of the product (Crowley et al., 2014). Das et al. (2015) emphasized the importance of density analysis to determine the type of packaging in the storage of starch samples. According to Santhalakshmy et al. (2015), Hausner ratio (HR) is used to evaluate the cohesiveness of the material, which can be classified as follows: HR < 1.2 - low cohesiveness, 1.2 < HR < 1.4 - intermediate cohesiveness, and HR > 1.4 - high cohesiveness. Thus, pitomba starch has high cohesiveness, which can be a desirable characteristic when it is applied to improve texture in food products, because cohesiveness is related to the tendency of the molecules to remain together. This characteristic could be preserved in the manufacturing of compositions.

The Carr index (CI) represents the flowability of a sample; CI between 15 and 20% indicates excellent flowability, between 20 and 35% indicates poor flowability, between 35 and 45% indicates very poor flowability and CI > 45 is attributed to materials with extremely poor flowability. The results for Carr index characterize pitomba starch as a product of poor flowability, which can be a desirable characteristic for starches used as food thickeners when one intends to make the flow difficult. Machado et al. (2012) analyzed the compressibility index of maize starch and reported CI of 36%, classifying it as a product of poor flowability.

Apparent viscosity
Figure 1 presents the apparent viscosity of pastes of pitomba starch at concentrations of 2, 4 and 6% under temperatures of 5, 15 and 25 °C, as a function of the rotation speeds. It can be observed that the pastes showed higher apparent viscosity as the starch concentration increased, but the values tended to decrease with increasing rotation speeds, i.e., due to the increase in shear stress, meaning that the samples behaved as a pseudoplastic fluid at all temperatures studied. According to Shamsudin et al. (2013), the increase of speed generates a force capable of causing structural change, which causes the particles to rearrange in parallel directions, leading to the break of large particles into smaller ones, and these can flow more easily in the direction of the applied stress. Li and Zhu (2018) report reduction of viscosity in pastes of quinoa starch gels with increase in shear rate, which is consistent with the findings of Zhu et al. (2016) and Kong et al. (2010) for starches of rice and amaranth, respectively.

By analyzing the behavior of viscosity as a function of temperature, it is possible to note a considerable reduction in the apparent viscosity of the pastes as the temperature increased. According to Pagno et al. (2014), temperature influences the apparent viscosity and higher viscosity is usually obtained under lower temperatures. Heymam et al. (2014), studying the rheological properties of pastes of modified maize starch, observed that their viscosity was higher at lower temperature, suggesting that the interaction between starch granules is higher under low temperatures because, despite being formed by microgel particles that do not undergo interpenetration, the starch granules overlap, becoming more viscous at lower temperatures. Silva et al. (2018) report higher viscosity in beverages formulated with different thickeners when these were stored under refrigeration temperature. The starch concentrations used (2, 4 and 6%) impart significant viscosity to the starch pastes, suggesting their application to reduce the flowability of liquids. According to Ai and Jane
(2015), the viscosity obtained in the paste determines the thickening power of the starch for various industrial applications, but the rheological characteristics of starch pastes depend on several factors, including starch chemical structure, starch concentration, temperature, shear rate and time.

**Syneresis**

The syneresis behavior of the ketchup formulations stored for 150 days at temperatures of 7 and 25 °C is shown in Figure 2. Storage time progressively influenced the evolution of syneresis in all ketchup formulations at both temperatures studied. The highest percentages of syneresis at all storage times and temperatures were observed in the C formulation (control), demonstrating the incapacity of the ketchup to preserve isotropy, maintaining the union of phases during storage, without the addition of a stabilizing ingredient in its composition. The formulations PB, MA and MB did not show syneresis immediately after processing at temperatures of 7 and 25 °C, demonstrating good water-binding capacity, forming elastic gels, a characteristic related to the starches added and their capacity to absorb water.

With addition of 0.5% of pitomba starch (PA), there was a reduction of syneresis in comparison to the control formulation (C), but there were percentages of syneresis immediately after processing, with progressive increase during storage. Arocas et al. (2009), using native starches of corn, waxy corn, potato and rice as thickening and stabilizing agents in white sauce, observed no syneresis in any of the formulations immediately after preparation. However, after being subjected to freeze/thaw cycles, syneresis was observed in sauces with 6% of corn and potato starches, and these authors point out that the occurrence of syneresis after processing the sauces is associated with structural changes.

The increase of pitomba starch concentration in the ketchup formulation imparted greater stability to the sauce. The addition of 1% of pitomba starch in the PB formulation was able to delay the syneresis, inhibiting its occurrence immediately after processing, but the progressive increase in syneresis during storage was also observed. According to Stahl et al. (2007), starch gels tend to form rigid structures during cooling, which means that the polysaccharide tends to retrograde as the storage time advances.

By comparing the efficiency of the types of starch used, it can be observed that the additions of 1% of pitomba and maize starches (PB and MB) had the same capacity to reduce syneresis during storage at both temperatures studied, being statistically similar for all storage times. The deceleration observed in the separation of water with the increase in starch concentration can be explained by the high capacity of the starch to absorb water during the gelatinization process.

In the comparison between the syneresis percentages of the formulations for temperatures of 7 and 25 °C (Figure 2A and 2B), higher values of syneresis were found in the ketchup at the higher temperature, which may be associated with the ease of reassociation of amylase and amylopectin chains under room temperature. A similar behavior was reported by Arocas et al. (2009), who studied syneresis of white sauce mixed with corn starch and found higher water release percentages at temperature of 20 °C.

**Apparent viscosity of the ketchup**

The behavior of the apparent viscosity of ketchups stored for 180 days under temperature of 7 °C is shown in Figure 3. In all formulations and storage times, the apparent viscosity of the samples decreased with the increase in deformation rate, an indication of the accommodative pattern of the structure of the ketchups, proportional to the increase in the applied shear stress, characteristic of the pseudoplastic behavior of the samples.

According to Mandala et al. (2004), the pseudoplastic behavior of starch-containing solutions occurs especially when the granules are significantly swollen, which leads to deformation when subjected to shear forces. In a comparison of the initial apparent viscosity values of the ketchup formulations between 0 and 30 days (Figure 3A and B), an elevation can be noted after storage under the temperature of 7 °C, which is related to the formation of a more rigid structure established by the retrogradation process, besides the effect of refrigeration on the increase in the firmness of pastes and sauces.

Increase of viscosity in the sauces at the initial times of storage was also observed by Sikora et al. (2007), studying the rheological and sensory properties of dessert sauces thickened by starch-xanthan gum combinations. These authors reported that the storage of sauces may result in a slow mutual organization of the ingredients which influence the structure of the food, causing changes in its viscosity.

After 30 days, all ketchup formulations showed a reduction in the initial viscosity. The addition of starch influenced the increase of viscosity, and alterations were observed from the addition of 0.5% of pitomba and maize starches. Diantom et al. (2017) also observed an increase in the apparent viscosity of tomato sauce with the addition of 0.5% of locust bean gum, potato protein, soy protein and potato fiber, which imparted higher flow resistance to the sauce. The highest initial apparent viscosities of the ketchups stored at 7 °C were observed in the formulations mixed with pitomba starch (PA and PB), possibly related to the composition of this starch, which has high content of amylopectin, with the ability to form more rigid gels due to the presence of a large number of branched chains that impart greater resistance to flow.

The behavior of the apparent viscosity of ketchups stored at 25 °C for 150 days is shown in Figure 4. Under this storage condition, the ketchups also showed pseudoplastic behavior, with reduction of apparent viscosity as the shear stress increased. Reduction of viscosity occurs due to the progressive orientation towards the flow, causing the disaggregation of the molecules formed from the sauce structure organization (Kim and You, 2009).

Increments in the initial apparent viscosity of all ketchup formulations were also observed after 30 days of storage, but at the temperature of 25 °C the ketchups showed greater increase of apparent viscosity in all formulations, compared to the samples at temperature of 7 °C, demonstrating lower influence of this temperature on the formation of rigid structures of pastes and sauces. According to Constenla et al. (1989), the reduction of viscosity with increasing temperature can be attributed to the increment in intermolecular distances in the starch, as a result of thermal expansion.
Table 1. Yield and physical-chemical characterization of pitomba starch.

| Parameters        | Result          |
|-------------------|-----------------|
| Yield (% d.b.)    | 44.20           |
| Moisture (% w.b.) | 8.41 ± 0.07     |
| Protein (%)       | 4.03 ± 0.05     |
| Ashes (%)         | 0.63 ± 0.05     |
| Lipids (%)        | 0.39 ± 0.02     |
| Starch (%)        | 77.78 ± 0.07    |
| Amylose (%)       | 19.25 ± 0.17    |

Figure 1. Apparent viscosity (mPa.s) of pitomba starch pastes at concentrations of 2% (A), 4% (B) and 6% (C) under different temperatures and rotation speeds.

Figure 2. Evolution of syneresis in ketchups stored for 150 days at 7 °C (A) and 25 °C (B). *Columns identified with identical letters at the same storage time do not differ statistically by Tukey test (p≤0.05) n=3.
Figure 3. Behavior of apparent viscosity in ketchups stored for 0 (A), 30 (B), 60 (C), 90 (D), 120 (E) and 150 (F) days at 7 °C.

Figure 4. Behavior of apparent viscosity in ketchups stored for 0 (A), 30 (B), 60 (C), 90 (D), 120 (E) and 150 (F) days at 25 °C.
Materials and methods

Pitomba fruits were purchased at the State Supply Center (CEASA) of the Recife city, Pernambuco, Brazil (8.071645 34.9425736,49W). The fruits were selected according to the yellowish-brown color of the peel and sent to the Laboratory of Storage and Processing of Agricultural Products of the Federal University of Campina Grande, Campina Grande, Paraíba, Brazil (7.21713155 35.9100279,97W).

Starch

To obtain native starch from pitomba, the fruit was manually peeled and the seed (endocarp), involved by the pulp, was immersed in sodium hydroxide solution (8%) for 30 min., followed by successive washings in running water to completely remove the pulp. Seeds resulting from the pulping process were neutralized in citric acid solution (4%) for 30 seconds. Starch extraction was carried out as described by Castro et al. (2019), through the immersion of seeds in sodium metabisulfite (0.2%) at 1:2 proportion (m/v) for 72 h. at 4 °C, followed by grinding of the mixture and sieving. The filtrate was left at rest for 1 hour, the supernatant was discarded, the starch paste was then washed in 0.2% metabisulfite solution, decanted (1 hour), and the supernatant was discarded twice. After decantation, the starch paste was centrifuged at 3000 rpm for 3 min. and the supernatant was discarded. The starch resulting from centrifugation was washed with alcohol (95%), centrifuged, with supernatant discarded 3 times, and dried in a forced air circulation oven at 50 °C for 12 h. The starch was ground in a domestic processor (Pratic Blender BLD300) to obtain the powder.

Yield, determination of moisture, crude protein, ashes, lipids, total starch and amylose

Starch extraction yield was evaluated based on the final mass of starch (d.b.) divided by the mass of endocarp used in the extraction (d.b.) and expressed in percentage. The contents of moisture, protein and ashes were determined according to the methods described by AOAC (2002): moisture in an oven at 105 ± 3 °C, procedure n° 925.09 (32.13); protein by Kjeldahl, using a conversion factor of 6.5, procedure n° 991.20 (33.2.11); ashes in a muffle furnace at 550 ± 3 °C, procedure n° 923.03 (32.1.05). Lipid content was determined by the method of Bligh and Dyer (1959). Starch content was evaluated according to the methodology proposed by the Adolfo Lutz Institute (IAL, 2004), whose principle is the detection of glucose produced by the hydrolysis of starch molecules, whereas amylose was quantified by the colorimetric method with results calculated based on a standard curve constructed with pure amylose (Sigma), according to the methodology of Zavareze (2009).

Table 2. Flow properties of pitomba starch.

| Parameters                 | Result |
|----------------------------|--------|
| Apparent density (g/cm³)   | 0.46   |
| Compact density (g/cm³)    | 0.66   |
| Hausner ratio              | 1.43   |
| Carr index (%)             | 30     |

Starch flow properties

Apparent density was determined according to Souza et al. (2010), by weighing 5 g of the pitomba starch sample in a 10-mL graduated cylinder, with no compaction, and calculated according to Equation 1. The starch contained in the cylinder was compacted by manually tapping the cylinder 50 times on a bench surface from a height of approximately 5 cm, to determine the compact density according to Equation 2. With these values of apparent and compact densities, the Hausner ratio and Carr index were calculated using Equations 3 and 4.

\[
\rho_a = \frac{m}{v} \quad (1)
\]

\[
\rho_c = \frac{m}{v_a} \quad (2)
\]

\[
HR = \frac{\rho_c}{\rho_a} \quad (3)
\]

\[
CI = \frac{\rho_c - \rho_a}{\rho_c} \quad (4)
\]

Where:

- \(\rho_a\) = apparent density (g.cm⁻³)
- \(m\) = mass (g);
- \(v\) = volume (cm³);
- \(\rho_c\) = compact density (g.cm⁻³)
- \(v_a\) = volume after compaction (cm³);
- HR = Hausner ratio;
- CI = Compressibility index (%)

Starch apparent viscosity

Solutions containing 2, 4 and 6% of starch were prepared, subjected to gelatinization in water bath (100 °C) for 30 min. and then cooled to room temperature. The cooled pastes were subjected to reading in a programmable viscosimeter DV-II+PRO, from Brookfield, at temperatures of 5, 15 and 25 °C, under rotation speeds of 20, 40, 50, 60, 80, 100, 120, 140, 150, 160, 180 and 200 rpm.

Preparation and storage of ketchups

Ketchups were prepared with tomato pulp (50%), water (25%), sugar (15%), vinegar (5%), salt (3%) and condiments (2%). Five formulations were prepared with different percentages of pitomba starch and maize starch (commercial) added to the standard formulation. Pitomba starch was subjected to a toxicity test according to Meyer et al., 1982, and presented DL₅₀ 1036.46 µg mL⁻¹, considered as non-toxic in compliance with Brazilian legislation for addition to food products. Ketchup formulations were identified as: PA - 0.5% pitomba starch, PB -
1% pitomba starch, MA - 0.5% maize starch, MB - 1% maize starch, and C - control.

After cooking, when the formulations reached the desired concentration (35 °Brix), the sauces were placed in 125-mL polypropylene plastic containers with lid and stored at temperatures of 7 and 25 °C for 150 days.

**Syneresis**

The stabilizing capacity of the starches in the ketchups was determined by quantifying the syneresis using the drainage method, by weighing 30 grams of the ketchup sample on a filter paper at the upper part of a funnel attached to a 100-mL Erlenmeyer flask, at 4 °C for 5 h. The index of syneresis was calculated according to Riener et al. (2010), using Equation 5:

\[
(\% \text{ Syneresis}) = \frac{\text{mass of water released in filtration}}{\text{initial mass}} \times 100
\]

**Apparent viscosity of ketchups**

The apparent viscosity of ketchups was evaluated to determine the thickening capacity of the starch in the formulations. A programmable viscosimeter (DV-II+PRO - Brookfield) was used at temperatures of 7 and 25 °C, with rotation speeds of 5, 10, 20, 40, 50, 60, 80, 100, 120, 140, 150, 160, 180 and 200 rpm.

**Statistical analysis**

The results were analyzed by descriptive statistics, using the measures of central tendency and data variability.

**Conclusions**

Pitomba starch showed low flowability and high cohesiveness, low percentages of lipids, ashes and protein. When gelatinized, the starch pastes showed plastic behavior and imparted good apparent viscosity, suggesting applications to increase the consistency of liquids, associated with a considerable reduction of viscosity with increasing temperatures.

Addition of pitomba starch to ketchup formulations led to higher apparent viscosity, compared to maize starch, and was able to delay syneresis in the sauces, which was increased with the increment in storage temperature.

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