

Effect of Tommy Atkins mango (*Mangifera Indica*) almond starch as a thickener in fish pâtés: Physicochemical and sensorial

Efeito do amido da amêndoa da manga Tommy Atkins (*Mangifera Indica*) como espessante em patês de pescado: Caracterização físico-química e sensorial

Efecto del almidón de mango y almendra Tommy Atkins (*Mangifera Indica*) como espesante en patés de pescado: Físicoquímica y sensorial

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Abstract

The aim of this study was to extract the starch from the Tommy Atkins mango (*Mangifera Indica*) and to evaluate the effect on the chemical and physico-chemical characteristics of tilapia pâtés. Starch was extracted and total starch, protein, moisture, ash, acidity, gelling, swelling and solubility, water and oil absorption capacity, and microbiological (thermotolerant coliforms, *Escherichia coli*, molds and yeasts, and *Staphylococcus aureus*) analyzes were performed. Pâtés were formulated with mango starch and corn starch, and the pH, lipids, moisture, ash, microbiological (Thermotolerant coliforms, *Escherichia Coli*, *Salmonella* sp., *Staphylococcus aureus*), instrumental color, sensory and stability characteristics were analyzed during storage. The extracted material presented 72.69% total starch, 2.96% protein, gelation in concentrations of 8 to 14%, greater swelling power and solubility at 95 °C, water retention capacity of 58 g/100 g and 45 g/100g of oil. The application of mango starch in pâté did not change its physico-chemical characteristics in comparison with pâté using corn starch. There were low counts of total coliforms, molds and yeasts in the starch and an absence of *Salmonella*, *Escherichia Coli*, *Staphylococcus Aureus* in the starch and pâtés in the microbiological analyzes. It was found that the pâté samples were stable for 30 days of cold storage, with low color and microbiological changes. The pâtés obtained good acceptance in the sensory evaluation with scores between 7 and 8. This study showed that the application of mango starch in tilapia pâté formulations is feasible, thereby constituting a means of reusing this residue.

Keywords: Agro-industrial waste; Stability; Tilapia; Sensory acceptance.

Resumo

O objetivo deste estudo foi extrair o amido da amêndoa da manga Tommy Atkins (*Mangifera Indica*) e avaliar o efeito nas características químicas e físico-químicas de patês de tilápia. Efetuou a extração do amido e analisaram-se amido total, proteínas, umidade, cinzas, acidez, geleificação, intumescimento e solubilidade, capacidade de absorção de água e óleo, e as análises microbiológicas (Coliformes termotolerantes, *Escherichia coli*, bolores e leveduras,

Staphylococcus aureus). Foram formulados patês com amido de manga e com amido de milho, e avaliou-se o pH, lipídeos, umidade, cinzas, microbiológicas (coliformes termotolerantes, *Escherichia coli*, *Salmonella sp.*, *Staphylococcus aureus*), cor instrumental, sensorial e estabilidade durante o armazenamento. O material extraído apresentou 72,69% de amido total, 2,96% de proteína, geleificação nas concentrações de 8 a 14%, maior poder de intumescimento e solubilidade a 95 °C, capacidade de retenção de água de 58 g/100g e de óleo de 45g/100g. A aplicação do amido de manga em patê não alterou suas características físico-químicas, em comparação com o patê utilizando amido de milho. Nas análises microbiológicas houve baixas contagens de coliformes totais, bolores e leveduras no amido e ausência de *Salmonella*, *Escherichia Coli*, *Staphylococcus Aureus* no amido e nos patês. Verificou-se que as amostras de patês mostraram-se estáveis durante 30 dias de armazenamento refrigerado, apresentando baixas alterações de cor e microbiológicas. Na avaliação sensorial os patês obtiveram uma boa aceitação com notas entre 7 e 8. Este estudo mostrou que a aplicação do amido de manga em formulações de patê de tilápia é viável configurando um meio de reaproveitamento desse resíduo.

Palavras-chave: Resíduos agroindustriais; Estabilidade; Tilápia; Aceitação sensorial.

Resumen

El objetivo de este estudio fue extraer el almidón del mango Tommy Atkins (*Mangifera Indica*) y evaluar el efecto sobre las características químicas y físico-químicas de los patés de tilapia. Se extrajo el almidón y se analizó el almidón total, proteínas, humedad, cenizas, acidez, gelificación, hinchamiento y solubilidad, capacidad de absorción de agua y aceite, y análisis microbiológicos (coliformes termotolerantes, *Escherichia coli*, mohos y levaduras, *Staphylococcus aureus*). Los patés fueron formulados con almidón de mango y almidón de maíz, y el pH, lípidos, humedad, ceniza, microbiológicos (Coliformes termotolerantes, *Escherichia coli*, *Salmonella sp.*, *Staphylococcus aureus*), color instrumental, sensorial y estabilidad durante el almacenamiento. El material extraído presentó 72,69% de almidón total, 2,96% de proteína, gelificación en concentraciones de 8 a 14%, mayor poder de hinchamiento y solubilidad a 95 ° C, capacidad de retención de agua de 58 g / 100 gy 45 g / 100 g de aceite. La aplicación de almidón de mango en paté no modificó sus características físico-químicas, en comparación con el paté que utiliza almidón de maíz. En los análisis microbiológicos, hubo recuentos bajos de coliformes totales, mohos y levaduras en el almidón y ausencia de *Salmonella*, *Escherichia Coli*, *Staphylococcus Aureus* en el almidón y patés. Se encontró que las muestras de paté se mantuvieron estables durante 30 días de almacenamiento en frío, con bajo color y cambios microbiológicos. En la evaluación sensorial, los patés obtuvieron una buena aceptación con grados entre 7 y 8. Este estudio mostró que la aplicación de almidón de mango en formulaciones de paté de tilapia es factible, configurando una forma de reutilización de este residuo.

Palabras clave: Residuos agroindustriales; Estabilidad; Tilapia; Aceptación sensorial.

1. Introduction

Mango (*Mangifera indica* L.) is a fruit belonging to the Anacardiaceae family. It has a large amount of pulp, is a variable size and shape, has a pleasant aroma and color to the consumer (Sarkar *et al.*, 2020). It is mainly consumed in natura due to its flavor, nutritional composition and beneficial health properties, as well as in the form of juices, nectars, jellies, and ready-made drinks, which have considerable demand for national and export markets (Lawson *et al.*, 2019). However, fruits with deficient quality are not accepted for commercialization during their industrialization, and so a large amount of waste is generated which comprises about 25 to 40% of fruits and unusable parts such as peels and seeds (Lehner & Siegmund, 2020). Therefore, these residues can be used in mango products, as well as to promote a reduction of their disposal into the environment (Bharti, Singh & Saxena, 2019).

It is possible to make flours from the green mango seed with a high lipid, fiber and starch content with a similar physico-chemical and functional composition to commercial starches, and can be used in several food products (Patiño-Rodríguez *et al.*, 2020). The starch obtained from *M. indica* seeds is considered one of the components to be used as a potential additive source, oxidative reaction stabilizer and edible coating on food (Ferreira *et al.*, 2019). Thus, these residues could be effectively used to produce starches for application in the food area to replace wheat flour, as mango seeds contain approximately 58% wheat flour (Lawson *et al.*, 2019; Nawab *et al.*, 2016). Some studies have partially replaced wheat flour with mango seed flour in formulating biscuits and also used the Totapuri variety mango seed flour as a source of phenolic compounds and partial fat substitute in breads (Menon, Majumdar & Ravi, 2015).

The food industry uses starches as basic ingredients in products to improve their manufacture, presentation and

preservation, since starches have thickening and gelling properties, in addition to being good texture regulators and stabilizers (Mahmood *et al.*, 2017). Starches can be added to the emulsion of meat and other food products due to their functional properties and their low costs (Genccelep *et al.*, 2017). Brazilian law allows the addition of starch into some meat products; one such product is pâté, defined as an industrialized product obtained from meat or edible mints from different species of butcher animals which are processed into paste, added to ingredients and subjected to a thermal process (Brasil, 2000). *Oreochromis niloticus* fish meats have been studied in the development of new products such as a creamy pâté, with the objective of encouraging fish consumption, adding value to the product and boosting commercialization, in addition to reducing the lack of this food in the market diet (Freitas *et al.*, 2012).

Given the above, the objective of this study was to extract and characterize the starch contained in the mango endocarp seed and use it in tilapia pâté formulations, thus contributing to the use of these agro-industrial residues and to investigate a new starch source for application in food.

2. Methodology

Plant material

The raw material used was the Tommy Atkins mango seed. The mangoes were purchased at a local supermarket, and the peels and pulp were manually removed using knives. The seeds were subsequently dehydrated in a Hareus B12 electric greenhouse (Hanau, Germany) at 50 °C for 24 hours, and then manually broken using a hammer and stainless steel knife, thereby obtaining the seeds.

Starch extraction

Starch isolation from the mango seeds was performed according to procedures described by Nawab *et al.* (2017) with adaptations. The seeds were cut into cubes and immersed in a 1% (w/v) sodium metabisulfite solution (Dinâmica, Brazil) for 12 hours. The water was then drained and the seeds were crushed with the addition of 1:4 (m/v) distilled water in a Waring industrial blender (USA) at 550 watts for 3 minutes until a thick and uniform mass was obtained. Next, the starch was filtered through 100 mesh nylon fabric, resuspended in distilled water and left to stand in a beaker to be decanted under cooling at 8 °C for 12 hours. After this time, the supernatant was eliminated and the starch was washed with distilled water and centrifuged at 3400 rpm for 15 minutes in a refrigerated centrifuge (Eppendorf f5804R, Hamburg, Germany). This procedure was repeated several times until the product was free of non-starchy residues. The obtained starch was transferred to beakers and dried in a vacuum oven at 45 °C for 14 hours and crushed in a blender (Mondial, Brazil) to obtain the powdered starch.

Physico-chemical characterization

Total starch was determined by spectrophotometry in a UV-Vis spectrophotometer (Shimadzu 3100) at 620 nm (IAL, 2008). Protein quantification was performed using Kjeldahl's total nitrogen determination method (AOAC, 2005). Humidity was determined in an oven at 105 °C until constant weight by the gravimetric method, ash by incineration in a muffle furnace at 550 °C and Total Titratable Acidity by titration (IAL, 2008). The results presented are the mean values of three replicates.

Gelling test

The method described by Lawal and Adebawale (2006) was applied the starch gelling test. Starch samples in concentrations of 2, 4, 6, 8, 10, 12, 14% (w/v) were prepared in test tubes with 5 ml of distilled water in triplicate, shaken for 5 min and the tubes were heated in a water bath with thermostat for 30 min at 80 °C, followed by cooling in cold running water. The tubes were then cooled to 4 °C for 2 h. The minimum gelling concentration was determined as that in which the inverted

tube sample did not slide through the tube.

Swelling and solubility

The swelling (SWP) and solubility (SOP) power of the starch were determined following the methodology described by Leach, Mccowen and Schoch (1959).

Water and oil absorption capacity

The water absorption capacity was performed according to the AACC (2000) by which 5 grams of starch was weighed in a centrifuge tube. About 30 mL of distilled water was added for complete hydration. The tubes with the samples were vortexed and centrifuged at 2000 rpm for 10 minutes. The supernatant was discarded and the tube weighed again. The oil absorption capacity was performed according to the method described by Beuchat (1977), in which 10 mL of soy oil (Soya, Brazil) was added to 1 g of starch sample in centrifuge tubes. The suspension was homogenized and left for 30 minutes. The tubes were subsequently centrifuged for 15 minutes at 3400 rpm. The result of the absorbed oil mass was expressed in g/100 g.

Microbiological analysis

The thermotolerant coliform (coliforms at 45 °C), *Escherichia coli* (EC), mold and yeast (MY) and DNA-positive *Staphylococcus* (STX) analyzes were performed using Petrifilm™ Plate kits (3M Microbiology, St. Paul, MN, USA) according to the AOAC 991.14, AOAC 997.02, and AOAC 2003.08 methods (AOAC International, 2003). The results presented are the mean values of three replicates.

Production of tilapia pâtés with mango starch added

Two formulations were defined: P1 - Pâté with mango starch added; and P2 - Pâté with corn starch added (both in the concentration of 4.2% starch). The proportions of the ingredients used were as follows: fish fillet 50%, water 24%, sodium chloride 1%, fish sauce 2%, color 0.32%, cream 18.5%, mango starch 4.2% (P1), corn starch 4.2% (P2). Tilapia fish (*Oreochromis niloticus*) fillets were thawed and cooked for 15 minutes at 74 °C to produce the pâtés. The ingredients were weighed on an analytical scale and the cooked fillet was crushed together with the other ingredients in a cutter (Metvisa, Brazil). The homogenized pâté mass was transferred to sterile glass jars and pasteurized in a water bath at 74 °C for 25 minutes, monitoring the temperature with an infrared digital thermometer (Smart SensorAR 360, Brazil). Next, the product was immersed in an ice bath for thermal shock until reaching the temperature of 8 °C. The pâté was stored in glass containers under refrigeration at 7 °C until the analysis.

Physico-chemical and microbiological characterization of pâtés

The physical-chemical analyzes of the pâté formulations were carried out by the methods previously mentioned for mango starch, as well as the pH analyzes using a pH meter (Even PHS-3E, Brazil), lipids by direct extraction in soxhlet, moisture and ashes according to IAL (2008). The microbiological characteristics were determined by analyzing thermotolerant coliforms (coliforms at 45 °C), *Escherichia coli* and DNA-positive *Staphylococcus* in Petrifilm™ plates (3M Microbiology, St. Paul, MN, USA) according to the AOAC 991.14, AOAC 997.02, and AOAC 2003.08 methods (AOAC, 2003), and *Salmonella* sp. according to APHA (2001). The results presented are the mean values of three replicates.

Color measurement

The color parameters on the surface of the pâtés were measured using a previously calibrated portable colorimeter

(Colorium 2 digital, Delta color, Brazil) with the Lab 7 software program according to CIELAB (AOAC, 2005), system The results were expressed with the color coordinates L* (Luminosity - L*=0 [black] and L* = 100 [white]), a* (green/red-when negative values indicate the degree of greening and when positive values indicate the degree of redness), b* (yellow/blue when -b* - blueness and + b* - yellowness), c* (croma - $c^* = (a^{*2} + b^{*2})^{0.5}$), h* (hue angle - $h^* = \arctg(b^*/a^*)$). Six readings were taken on each sample.

Stability analysis of tilapia pâtés

pH analyzes were performed according to the methodology described by IAL (2008) and the microbiological analyzes of total coliforms (35 °C), *Escherichia coli* (EC), and *Staphylococcus aureus* (STX) using petrifilm™ plates (3M Company, St. Paul, MN, USA), as well as Salmonella sp. (APHA, 2001) to evaluate the microbiological stability of pâtés produced on days 0, 14 and 30. Furthermore, lipid oxidation was determined following the methodology described by Silva *et al.* (2003), in which the amount of substances reactive to 2-thiobarbituric acid (TBARS) was verified using tetramethoxypropane (TMP, Sigma ChemicalCo.) as standard and the values were expressed in mg of malonaldehyde per kg of sample. In addition, the color parameters of the sample were checked using a Delta Color colorimeter according to the CIELAB system (AOAC, 2005). The parameters: L, a*, b*, c* and h* were evaluated by taking six readings in each treatment (P1 and P2), at different storage times (0, 14 and 30 days). Six readings were taken on each sample.

Sensory analysis of pâtés

A sensory analysis with an untrained panel of 90 tasters aged over 18 years was conducted. The preference/acceptance test was applied through the nine-point hedonic scale test, ranging from 1 (very disliked) to 9 (very much liked) (Stone & Sidel, 2004) for the attributes of color, aroma, texture, flavor and overall acceptance. The pâtés were spread on toast and served to the tasters in plastic dishes encoded with random numbers of three digits at refrigerated temperature (8 °C ± 1), accompanied by a glass of water to remove the residual flavor between tasting the samples. The project was approved by the ethics committee of the Federal Institute of Education, Science and Technology of Ceará with the opinion number 2.586.703.

Statistical analysis

The data obtained for the starches and the different pâté treatments were evaluated through analysis of variance (ANOVA) followed by a comparison between the pairs of means by the Tukey test ($p < 0.05$). The statistical software used for this purpose was the Excel 2.9 Action supplement (Statcamp, São Carlos, Brazil).

3. Results and Discussion

Physico-chemical characterization

The results of the characterization of mango seed starch are shown in Table 1. The moisture and ash parameters showed values of 18.30 and 0.99, respectively, therefore above that allowed by the legislation for commercial starches which limits the maximum allowed to 13-15% for moisture and 0.5% for ash. High moisture values can be justified due to the drying method used and possible inadequate conditions; therefore, they can be solved by using other methods of drying the material until obtaining lower water content. Another justification for the high moisture content may be due to the prior lack of drying the raw material used (Silva *et al.*, 2013).

Table 1 - Mean results of the physicochemical characterization of mango starch.

Physico-chemical parameters	(%)
Moisture	18.30 ± 0.36
Ashes	0.99 ± 0.12
Proteins	2.96 ± 0.03
Total Starch	72.69 ± 0.11
Total Titratable Acidity *	0.30 ± 0.04

* Citric acid (g/100 g). Source: Authors.

Ferreira *et al.* (2019) found moisture values for mango seed starch obtained by atomization ranging from 6.41% to 3.53%. Regarding the amount of ash, studies on the characterization of mango starch reported ash values of 0.35% lower than those found in this study (Silva *et al.*, 2013). There was a value of 0.30 g/100 g of citric acid, and this lower result may be related to the formation of the amylose-lipid complex which favors reducing titratable acidity (Ferreira *et al.*, 2019).

The protein content quantified in the starch was 2.96%. There may have been a denaturation of proteins during the drying process (Bharti *et al.*, 2019). This result is satisfactory, since it is advisable to have low protein levels adhered to the starch granules, since reduced protein levels indicate high starch purity (Guo *et al.*, 2018). Silva *et al.* (2013) reported a 2.33% protein value for starch in evaluating the use of starch extracted from the Tommy Atkins mango seed and its application as a thickener in a dairy drink. Starch is practically composed of carbohydrates, however it contains substances such as lipids, proteins and ash, and its amount in starch depends on the plant composition and the extraction and purification methods (Cavalcanti *et al.*, 2011).

The starch content quantification presented 72.69%. This value is higher than those found in work developed with starch extracted from the Chaunsa mango variety of 56% (Nawab *et al.*, 2016), and those reported by Fernandes *et al.* (2019) for starch from Tommy Atkins mangoes which ranged from 58.4% to 69.8%. On the other hand, this value is in accordance with the studies by Patiño-Rodríguez *et al.* (2020), who reported values of 74% for starches made from green mangoes.

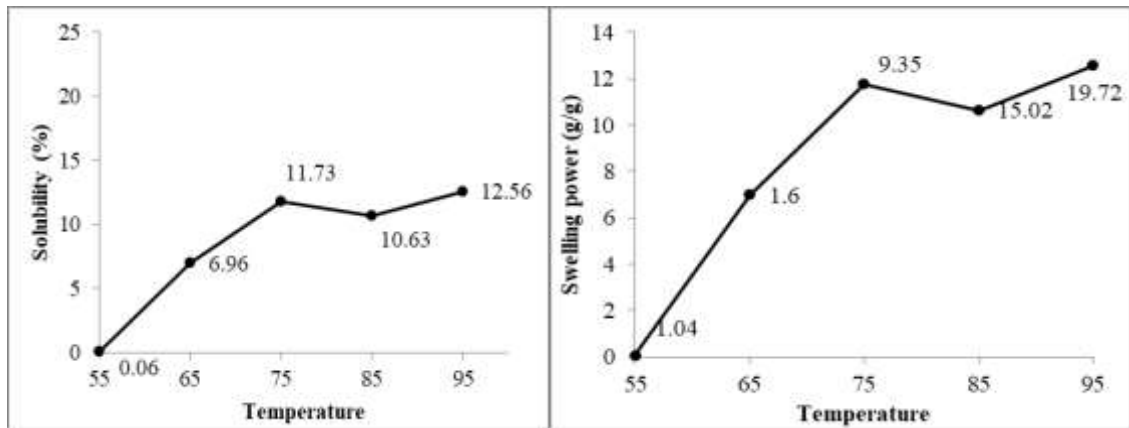
Starch gelling

It was found that the minimum concentration for gel formation in the studied starch was 8%, with no gelatinization in concentrations of 2, 4 and 6% (liquid state) and positive in concentrations of 8 to 14% with the formation of viscous to very firm gel. Positive gelation in all concentrations with minimum gelation in the concentration of 6% was obtained in studies on the gelation of commercial corn starch in the same concentrations (Cavalcanti *et al.*, 2011). It is possible to verify that the mango seed starch to be used as a thickener in the food industry needs better purification in order to present satisfactory physico-chemical characteristics similar to commercial corn starch, however a similar gelling property is achieved, showing that this residue can be used to develop a new ingredient which has not yet been commercially exploited.

Swelling and solubility power

The water solubility and swelling of mango starches are shown in Figure 1. The swelling of the starch is related to its ability to absorb water and the solubility is related to the degree of dissolution of the starch, both during heating (Guo *et al.*, 2018). It can be seen that the swelling power (SWP) fluctuated from 1.04 g/g to 19.71 g/g.

Figure 1. Effect of temperature variation on the Swelling Power (SWP) and Solubility (SOP) of mango seed starch.



Source: Authors.

It was observed that as the temperature rises, the SWP increases in the same proportion, and the analyzed starch reached the maximum swelling value at 95 °C. The reduced values for SwP were possibly influenced by the structure of the amylopectin chains (He & Wei, 2017).

The swelling capacity of different starch granules not only depends on their binding to water, but also on several other factors such as starch concentration, amylose content, temperature and particle size, stiffness and susceptibility to molecular disentanglement, protein content and lipids (Bharti *et al.*, 2019; Guo *et al.*, 2018).

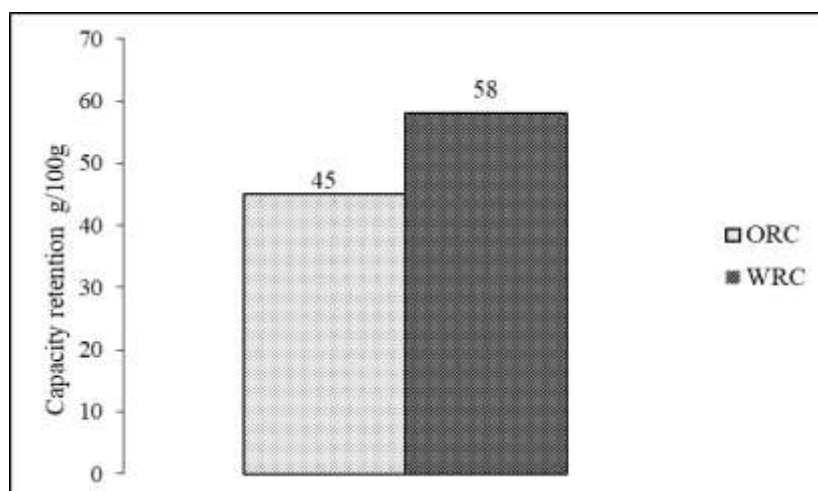
Studies on kernel starch from five Indian mango varieties showed SWP values between 18.0 and 19.7 (g/g), and that the low SWP can vary due to the presence of lipids in the starch which forms a complex with amylose (Nawab *et al.*, 2016). Siroha *et al.* (2020) found swelling values of 14.8%, being close to those reported in this study for starches made from mango seeds. The authors revealed that this parameter is related to the proportion of amylose and amylopectin and how they are arranged in the starch granule.

The solubility values quantified for mango starch ranged from 0.06% to 12.56%, in which the maximum solubility power (PS) was reached at 95 °C. In research carried out on the properties of starches extracted from two mango varieties, the authors observed that the solubility of the criollo variety reached a maximum solubility of 15%, while the manilla reached 26%, both at 80 °C. Nawab *et al.* (2017) studied the application of mango starch as coatings and reported a solubility of 8.71% at 90 °C. The variations in the solubility power found between the different starches may be due to differences in the length and distribution of the amylose chain.

Water and oil retention capacity

The water retention capacity (WRC) of mango seed starch was 58 g/100 g (Figure 2), which is less than what is reported in other scientific studies.

Figure 2. Mango seed oil and water retention capacity.



Source: Authors.

These reduced values are possibly related to the high moisture of the evaluated starches (Bharti *et al.*, 2019). Nawab *et al.* (2016) quantified the WRC of 77 g/100 g in research on the properties of mango seed starch. It was possible to observe that the water retention capacity of mango starch was greater than the oil retention. It is suggested that this starch may be suitable to be used as an ingredient for the preparation of pâtés, since the quality of the meat emulsion in this type of product is associated with good water retention (Ordóñez, 2005).

The results found in the present work are lower than the WRC of starches from conventional sources, for example, the WRC of corn starch can vary from 180 g/100 g to 208 g/100 g (Singh *et al.*, 2014). Furthermore, a value of 45 g/100 g was observed regarding the oil retention capacity (ORC) found in this work. The oil retention capacity of the mango starch studied was higher than that of rice and corn starch found in a study carried out in India with values of 1.09 g/100 g - 1.10 g/100 g for rice starch, and 0.80 g/100 g to 0.85 g/g for corn starch (Singh *et al.*, 2014).

Physico-chemical characterization of Nile tilapia pâté

The mean results for the physico-chemical characterization of tilapia and mango starch pâtés are shown in Table 2.

Table 2. Results of physico-chemical characterization of tilapia and mango starch pâtés.

Parameters	P1 (g/100g)	P2 (g/100g)
Moisture	68.09 ± 0.63 ^a	67.82 ± 1.16 ^a
Ashes	1.78 ± 0.02 ^a	1.81 ± 0.03 ^a
Lipids	14.18 ± 0.94 ^a	15.31 ± 0.13 ^a
pH	6.51 ± 0.05 ^a	6.42 ± 0.09 ^a

Distinctive superscript letters in the same column indicate a significant difference ($p < 0.05$) between the different treatments. P1: Pâté with mango starch added; P2: Pâté with corn starch added. Source: Authors.

It can be verified that there was no significant differences ($p > 0.05$) in any of the physicochemical parameters evaluated when comparing the tilapia pâté produced with mango starch (P1) and the pâté produced with corn starch (P2) (Table 2). The quantified values of moisture and lipids in both products are in accordance with the standards established by the technical regulation of identity and quality of pâté, which allows a maximum of 70 g/100 g of moisture and 32 g/100 g of

lipids. There is no standard in the current legislation regarding the ash content which applies to the preparation of pâtés. However, the quantification of this parameter is useful to estimate whether a given food is a good source of minerals. The two pâté formulations had low ash content.

The pH of the formulations was between 6.51 and 6.42 for mango and corn starches, respectively, being in accordance with results reported in related scientific works, which highlight that the pH of pâtés can vary from 6.54 to 6.57 (Delgado-Pando *et al.*, 2011). These values close to neutrality are common in fish pâtés, since the pH of fish meat is higher than other meats due to the lower glycogen content providing a higher pH after slaughter (Ferreira *et al.*, 2016).

Microbiological analysis of mango starch and tilapia pâtés

The results of counting total coliforms in mango starch and elaborated pâtés ranged from 3.0×10^{-2} to 6.0×10^{-2} (CFU/g), with the absence of *Escherichia coli*, *Salmonella* and *Staphylococcus aureus* also being observed (Table 3).

Table 3. Results of microbiological analyzes for mango starch and prepared pâtés.

Parameters	MS	P1	P2
Total coliforms (CFU/g)	6.0×10^{-2}	3.0×10^{-2}	6.0×10^{-2}
<i>Escherichia coli</i> (CFU/g)	Absence	Absence	Absence
Molds and yeasts (CFU/g)	3.0×10^{-2}	-	-
<i>Staphylococcus aureus</i>	Absence	Absence	Absence
<i>Salmonella sp.</i> $25g^{-1}$	Absence	Absence	Absence

MS: Mango starch; P1: Pâté containing mango starch; P2: Pâté containing corn starch. Source: Authors.

A value of 3.0×10^{-2} CFU/g was observed in quantifying the molds and yeasts from mango starch. Microbiological analyzes showed satisfactory values such as low counts of total coliforms, molds and yeasts, and an absence of *Escherichia coli*, *Salmonella* and *Staphylococcus aureus* for both mango starch and pâtés, and therefore they were within the standards established by legislation representing a microbiologically reliable product to be used as an ingredient in food formulations fit for human consumption.

Color measurement

Table 4 shows that there was a significant difference ($p \leq 0.05$) for all colorimetric parameters evaluated in the different pâté treatments.

Table 4 - Results of measuring instrumental color in the elaborated pâtés.

Treatments	Colorimetric coordinates				
	L*	a*	b*	c*	h*
P1	42.72 ± 0.01^a	1.82 ± 0.01^a	8.49 ± 0.01^a	8.68 ± 0.01^a	77.93 ± 0.06^b
P2	42.04 ± 0.01^b	1.49 ± 0.01^b	8.17 ± 0.01^b	8.30 ± 0.02^b	79.64 ± 0.06^a

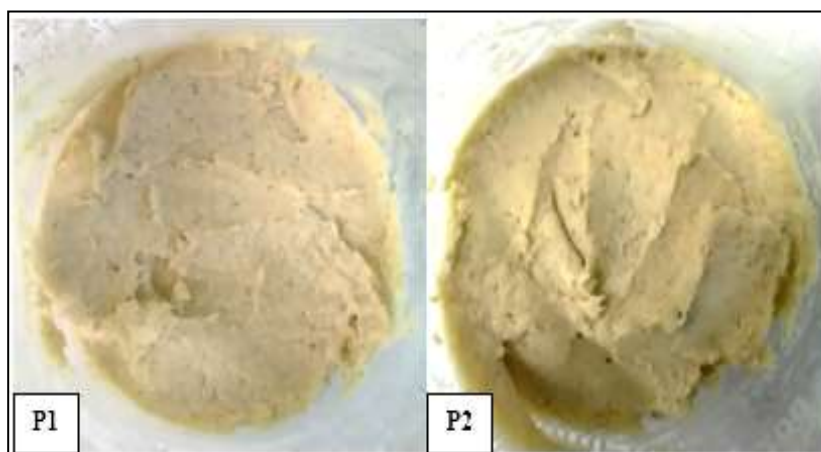
Distinctive superscript letters in the same column indicate a significant difference of 5% ($p < 0.05$) between the different treatments. P1: Pâté with mango starch added; P2: Pâté with corn starch added. Source: Authors.

Pâtés made with the addition of mango starch (P1) had a higher luminosity value L* compared to those with corn

starch added (P2) (Table 4). The L* coordinate is influenced by the water retention capacity, fat content and free water on the product surface (Sánchez-Zapata *et al.*, 2011). These results may be related to the low water retention of mango starch (Fig. 2), thus it is suggested that P1 had a greater amount of free water on the product surface, influencing the values obtained for luminosity. On the other hand, the figures suggest that the two formulations were opaque and less shiny. The averages found regarding the a* parameter which indicates red to green tint for the two formulations indicate a color with a tendency to dark green; however, the P1 sample showed a slightly higher average compared to sample P2.

All samples showed positive values for the b* parameter, meaning that a yellowish appearance is evident for both samples, but parameter b* was higher in the P1 treatment. The values obtained for c* and h* confirmed the yellowish tendency of the samples, however the color intensity, which is the highest saturation c* value, was observed in the sample with mango starch added (P1) (Figure 3).

Figure 3. Pâté samples made with mango and corn starches.



Source: Authors.

Evaluation of the stability of pâtés

Table 5 shows the mean values and standard deviation of the pH and TBARS parameters of the pâtés at different storage times. The pH values for the two formulations of tilapia pâté ranged from 6.82 to 5.66 during the 30 days of storage. It can be observed that there was a significant difference ($p \leq 0.05$) between treatments and storage time. Regarding treatments, it was observed that there was a significant difference in pH values only on the 30th day, with formulation P1 having the highest value (6.82).

Table 5. pH and TBARS values of pâtés at different storage times.

Parameters	(Days)	pâtés	
		P1*	P2*
pH	0	6.30±0.01 ^{aA}	6.30 ± 0.28 ^{aC}
	14	6.26±0.09 ^{aA}	6.48 ± 0.01 ^{aB}
	30	5.66±0.38 ^{bB}	6.82 ± 0.02 ^{aA}
TBARS (mg de malonaldehide /Kg)	0	0.007±0.03 ^{bC}	0.008 ± 0.02 ^{bC}
	14	0.018±0.04 ^{bB}	0.022 ± 0.03 ^{bB}
	30	0.52±0.02 ^{aA}	0.57 ± 0.02 ^{aA}

* P1: pâté with mango starch and P2: pâté with corn starch. a, b Distinctive superscript letters on the same line indicate a significant 5% difference ($p \leq 0.05$) between the different treatments for the same day of storage. A, B Distinctive superscript capital letters in the same column indicate a significant difference of 5% ($p < 0.05$) between different storage days for the same treatment. Source: Authors.

This increase in pH is suggested for protein degradation, and consequently the formation of substances such as ammonia and amines (Kirschnik & Macedo-Viegas, 2009). According to Brasil (2017), the pH of the fish meat both internally and externally must not exceed 7.0, therefore, the pH values of the pâtés were close to neutrality as 50% in the samples was based on a tilapia filet. With regard to time, there was an increase in the mean pH values for P1, with a statistical difference at 5% probability. On the other hand, there was a reduction of these values in the sample with mango starch (P2) over time; this fact is possibly be related to the development of lactic bacteria during storage (Ferreira *et al.*, 2016).

There was also a statistical difference ($p \leq 0.05$) for TBARS between the samples and the storage time (Table 5). The values during the storage times varied from 0.007 to 0.57 mgMDA/Kg. An increase in the values can be observed in the two samples, with a significant difference on the last day of storage. However, these increases were not sufficient for the production of compounds responsible for lipid oxidation (Ferreira *et al.*, 2016). According to the limits endorsed by the scientific literature, values above 1-2 mg of malonaldehyde/kg of fish are associated with the characteristic rancid odor and flavor (Tomita *et al.*, 2006).

Table 6 shows the averages of the color parameters of the pâtés evaluated at different storage times.

Table 6 - The mean \pm standard deviation L*, a*, b*, c* and h* values of the pâtés evaluated for the different storage times.

Parameters	Period (Days)	Instrumental Color	
		Pâtés	
		P1*	P2*
L*	0	80.56 \pm 0.23 ^{Aa}	84.15 \pm 0.22 ^{Aa}
	14	77.42 \pm 0.02 ^{Bb}	75.04 \pm 0.05 ^{Bb}
	30	72.57 \pm 0.22 ^{Bc}	70.55 \pm 0.35 ^{Bc}
a*	0	16.00 \pm 0.09 ^{Aa}	8.46 \pm 0.48 ^{Bc}
	14	14.43 \pm 0.22 ^{Ab}	15.05 \pm 0.05 ^{Ab}
	30	11.77 \pm 0.21 ^{Bc}	19.42 \pm 0.10 ^{Aa}
b*	0	52.74 \pm 0.48 ^{Ba}	55.88 \pm 0.28 ^{Aa}
	14	50.19 \pm 0.15 ^{Bb}	53.37 \pm 0.31 ^{Ab}
	30	47.74 \pm 0.24 ^{Bc}	50.55 \pm 0.32 ^{Ac}
c*	0	55.08 \pm 0.38 ^{Aa}	56.61 \pm 0.21 ^{Aa}
	14	52.09 \pm 0.07 ^{Bb}	55.31 \pm 0.12 ^{Ab}
	30	50.89 \pm 0.08 ^{Bc}	53.25 \pm 0.18 ^{Bc}
h*	0	73.27 \pm 0.45 ^{Bc}	81.59 \pm 0.51 ^{Aa}
	14	74.22 \pm 0.07 ^{Ab}	74.26 \pm 0.04 ^{Bb}
	30	75.26 \pm 0.01 ^{Aa}	70.74 \pm 0.13 ^{Bc}

* P1 - pâté with mango starch added and P2 - pâté with corn starch added. a, b: Different superscript lower case letters on the same line indicate a significant difference of 5% ($p \leq 0.05$) between the different treatments for the same day of storage. A, B: Different superscript capital letters in the same column indicate a significant difference at 5% ($p \leq 0.05$) between the different days of storage for the same treatment. Source: Authors.

It was possible to observe that there was a significant difference ($p \leq 0.05$) between treatments and the time for the color parameters evaluated in the three storage times. High luminosity values (L*) were found for both formulations, indicating that the samples were clear. However, a gradual reduction in the values was observed in the two samples evaluated, which is most notable in the pâté based on corn starch (P2), causing darkening of the samples, which may be related to oxidative

processes during storage (Ferreira *et al.*, 2016). P1 tended to have a greater appearance of red compared to P2 in relation to the a^* parameter at the beginning of storage, however it decreased on the 30th day. On the other hand, there was an increase in this color during storage for the P2 treatment, presenting higher values on the last day (19.42).

All samples showed positive values for the b^* parameter, meaning a tendency towards yellow color, and which was evident for both samples; however, the b^* parameter was higher in treatment P2, possibly due to the greater water retention capacity of corn starch. However, there was a reduction in these values, which shows that the samples became less yellow during storage.

The results of a^* and b^* parameters may be related to the incorporation of colorophilic in the formulation of pâtés, intensifying the red and yellow color of the samples. The main pigment in the coloroficle which has annatto as its main pigment is bixin, which represents about 80% of the total carotenoids, whereas the powder extracted from the seed is widely used in the food industry as a condiment to enhance the food color (Franco *et al.*, 2002).

The samples were statistically equal for the chroma, with a significant difference ($p \leq 0.05$) only in relation to the storage time. Thus, the two treatments showed similar color intensity during the study; however, there was a reduction in the values in both formulations, being higher in the sample P1. Therefore, the color intensity of the evaluated pâtés was altered with storage, possibly caused by pigment oxidation.

There was a statistical difference ($p \leq 0.05$) for the hue angle (hue) between treatments and time, with reduced values in both formulations during the 30-day storage period. According to the results, the samples showed a tendency to red color, which is justified due to the addition of ingredients such as seasonings and coloring in the formulations. On the other hand, there was a reduction in values during storage, in which the formulation with mango starch (P1) showed less color change compared to the sample with corn starch. This change in hue is possibly related to the degradation of pigments and chemical reactions, which can be influenced by the sample composition, pH and temperature, in addition to reactions such as Maillard's caused by the protein-aldehyde interaction (Nuanmano; Prodpran & Benjakul, 2015).

Microbiological stability

The results of the microbiological evaluation for the total coliforms (35°), *Escherichia coli* (EC), *Staphylococcus aureus* and *Salmonella* sp. for the pâté samples in the storage period (0, 14 and 30 days) indicated the presence of total coliforms (35 °C), with values of 27×10^{-1} CFU/g on the last day of storage (30th day) for the pâté made with mango starch (P2). The results for *Escherichia coli* (EC), *Staphylococcus aureus* and *Salmonella* sp. were absent in the two pâté treatments at all different storage times. This shows that the tilapia pâtés were in good condition for human consumption, taking into account the different storage times evaluated in the present study.

Sensory evaluation

Table 7 shows the results of the sensory acceptance of pâté samples based on tilapia filets made with mango and corn starch.

Table 7 - Result of the sensory acceptance of tilapia pâtés made with mango and corn starches.

Sensory attributes	Treatments	
	P1	P2
Color	7.80 ± 1.00 ^a	7.83 ± 0.98 ^a
Aroma	7.72 ± 1.05 ^a	7.92 ± 0.95 ^a
Texture	7.72 ± 1.29 ^a	8.00 ± 0.86 ^a
Flavor	7.94 ± 1.17 ^a	8.32 ± 0.80 ^b
Overall impression	8.02 ± 0.94 ^a	8.16 ± 0.72 ^a

Distinctive superscript letters in the same column indicate a significant difference of 5% ($p \leq 0.05$) between the different treatments. P1: Pâté with mango starch added; P2: Pâté with corn starch added. Source: Authors.

The sensory attributes evaluated for the two pâté formulations obtained averages which ranged from 7.72 to 8.32 corresponding to the hedonic terms “I liked it moderately” and “I liked it a lot”, which shows that the two samples obtained sensory acceptance from some of the tasters (Table 7). These results are superior to those reported in studies on the sensorial acceptability of tilapia meat pâté, which presented values ranging from 6.99 to 7.56 (Freitas *et al.*, 2012).

According to the results, it was observed that there were no statistical differences ($p > 0.05$) between the formulations, except for the flavor attribute ($p \leq 0.05$), since the pâté with corn starch added (P2) obtained a slightly elevated mean (8.32) compared to that made from mango starch (P1). These results may be related to the accentuated salt flavor of the P1 sample, as some tasters described the sample as saltier, thus a significant effect was obtained on the flavor attribute; however, the pâtés were well accepted by tasters according to the averages found. There was no statistical difference between the two formulations regarding the overall impression, which presented averages between 8.02 and 8.16 for the assigned scores. Therefore, the addition of mango starch to tilapia pâtés did not alter most of the evaluated sensory characteristics, which shows that this type of starch can be an alternative as a substitute for commercial starch.

4. Conclusion

The application of seed starch from the mango endocarp in tilapia pâté formulations is feasible, configuring a means of using agro-industrial residues. The endocarp seeds of the Tommy Atkins mango showed a high starch concentration, showing good gelling properties. The use of starch in pâté formulations did not significantly affect the physical-chemical parameters, nor did it alter their microbiological stability. In addition, the prepared pâtés showed good sensory acceptance and stability over 30 days of storage. As suggestions for future work, it is possible to carry out the study of mango starch as thickeners in other food products, as well as to evaluate its rheological capacity and antioxidant activity.

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