Physicochemical and sensory assessment of partial corn substitutions with carotenoid-containing non-traditional flours during tortilla preparation

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Abstract: Tortilla is a staple food for several countries and a gluten-free alternative and can be enriched using other non-traditional flours to improve their nutritional value. Three different lime and moisture concentrations were tested, and 0.25 and 56.34 g/100 g were selected as the best parameters to formulate during the rest of the survey. Once these conditions were determined, three different flours of sweet potato, peach palm, and cassava, at levels of 10 and 25 g/100 g, were partially used to substitute nixtamalized corn during the tortilla preparation. Several nutritional parameters of the flours were determined, including amino acid, mineral, fatty acid, sugar profiling as well as dietary fiber (sweet potato ≈ corn > cassava > peach palm), and carotenoids (peach palm > sweet potato > cassava ≈ corn). Moisture behavior and capacity of rollability during tortillas storage were also monitored for 15 days at 4 °C. Of all substitutions, only with sweet potato substitutions and the cassava substitution at a 10 g/100 g level, the typical puffing for a traditional tortilla is observed. Meanwhile, substitutions using peach palm and cassava flour showed the most degradation in storage. Sweet potato and peach palm (60.0 and 58.8 g/100 g) are less starchy ingredients than corn and cassava (87.8 and 85.30 g/100 g). Also, resistant starch for the tortillas ranged from 0.945 to 1.336 (cassava > peach palm > sweet potato), values higher than those found in a corn tortilla. Sensory analysis was also performed; both sweet potato and peach palm substitutions at 25 g/100 g obtained the lowest values of approval, by a consumer panel, for color and taste, respectively. Rollability was diminished as refrigeration time progressed, but sweet potato tortilla seems to withstand degradation (4.67–5.00 rollability scale) more than the control or other treatments (i.e., < 4). Overall, the partial substitution of corn flour improved the resistance to shear and deformation; this was especially true for the sweet potato substitution at day 0. Sweet potato substitutions generated a tortilla with a significantly higher induction period (ca. 45 hours, p < 0.05) and calculated shelf life (ca. 5 hours) than the other treatments. It can be demonstrated that using non-traditional flours can improve several tortilla parameters and be an additional vehicle for carotenoids in the diet.

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Keywords: corn tortilla; alternative starch sources; supplementation; sensory and chemical analysis; storage stability

1. Introduction
Tortilla, a thin, round, unleavened, and flattened bread with diameters ranging from 160 to 300 mm and 2 to 6 mm thick (Guzmán-Soria et al., 2019), is a staple food in Mexico and throughout Central-American countries, and it is popular in the United States (Guzmán-Soria et al., 2019). Tortillas are in great demand as the North, and Central American market was projected to reach USD 29.39 billion over the next three years (Guzmán-Soria et al., 2019); it can be considered a globalized product (Gabel, 2003).

In Mexican urban and rural environments, the consumption of tortillas is calculated at 56.7 and 79.5 kg per capita per year (Guzmán-Soria et al., 2019). Meanwhile, tortilla consumption in the United States and Europe rounds up to 6 (which represents 120 million tortillas per year) and 0.3 kg per capita per year, respectively (Rooney & Serna-Saldívar, 2016; Serna Saldívar & Chuck-Hernández, 2016), making these the second most popular baked product, after white bread. Hence, the economic and cultural importance of the tortilla is evident (Guzmán-Soria et al., 2019).

Corn is the second largest crop worldwide, producing 615,533,645 Mton (millions of tons; Mendoza-Cano et al., 2016). This grain has many food uses, among which is its use for making tortillas, which deliver fiber, whole grains, and other nutrients while being lower in fat and calories than wheat flour tortillas. Nutritionally, tortillas contribute ca. 37, 39, and 45% of total calories, protein, and calcium, respectively (Cruz-Huerta & Verdalet-Guzmán, 2007).

Corn tortillas are obtained through a process known as nixtamalization, in which the corn is cooked in an alkaline medium. Then, the wet grain is grounded to break its external structure and disperse the cellular components in addition to the starch polymers. A mass is obtained, a network of starch polymers solubilized together with scattered, raw, and swollen starch granules, cell fragments, proteins, and lipids (Quintanar Guzman et al., 2009).

When it is cooked, the gelatinization process begins, where amylpectin and amylase are hydrated, which causes the starch granule to break up and form a kind of gel. When the temperature decreases, the phenomenon of retrogradation can be observed, where there is a rearrangement of the structures of the starch present in the gel, which causes water loss, so that essential properties such as texture and useful life are affected (Rendón-Villalobos, Agama-Acevedo, Islas-Hernández, Sánchez-Muñoz et al., 2006b).

To improve the texture and durability of tortillas produced industrially, certain additives such as gums or hydrocolloids will enhance the tortillas’ texture characteristics, give them greater flexibility and maintain their rolling capability even during storage (Platt–Lucero et al., 2010). Additionally, despite already being an alimentary relevant product, corn tortillas’ overall nutritional and physical characteristics may be improved by adding natural products (e.g., Lys and Trp fortification; Leucona-Villanueva et al., 2012).

Given the above, it is of general interest to study the combined effect of adding hydrogels and partial substitution of corn flour by alternative starch sources. Adding or partially substituting other vegetable flours for tortilla products can diversify the variety of available gluten-free foods (Hosseini et al., 2018). Furthermore, hydrogels have already been demonstrated to improve dried nixtamalized maize masa’s thermal, functional and rheological properties (Aguirre-Cruz et al., 2005).

Some studies have already analyzed rheological and sensory aspects of other types of composites and partial substitutions of corn during tortilla preparation, with acceptable sensory
characteristics and improved moisture absorption resistance, including chia seeds (Rendon-Villalobos et al., 2012), β-glucans (Sanchez-Madrigal et al., 2015), malanga (Chel-Guerrero et al., 2015), banana (Aparicio-Seguiian et al., 2013), beans (Anton et al., 2009, 2008; Treviño-Mejia et al., 2016), amaranth (Vázquez-Rodriguez et al., 2013), distillers dried grains with solubles (Pourafshar et al., 2015), triticale (Hussein et al., 2013; Vaca-Garcia et al., 2011), wheat (Bejosano et al., 2005), jumbo squid muscle (Heredia-Sandoval et al., 2021), broccoli (Vázquez-Durán et al., 2014), algae (Quintero et al., 2014), and even non-toxic physic nut (Jatropha curcas L.; Argüello-Garcia et al., 2017).

Despite these modifications to tortilla formulations, non-traditional flours have seldom been addressed as an additional carotenoid source. The objective of the study was to incorporate carotenoids (considering their potential health benefits, Kumar Saini et al., 2022) into traditional corn tortillas and evaluate whether the senso-physicochemical characteristics of the tortillas were preserved or even could be improved and vary the classic flavor of the tortilla but also extend the shelf life of the product, but always keeping this gluten-free alternative. To this end, tortillas were prepared using three carotenoid-rich non-traditional, inexpensive, and readily available tropical flours, i.e., cassava (sweet yellow/cream pulp varietal, Manihot esculenta Crantz), sweet potato (Ipomoea batatas (L.) Lam.), and peach palm (Bactris gasipaes Kunth) at two different levels, each, in conjunction with two hydrogels (i.e., carboxymethyl cellulose and guar gum) as partial substitution of corn.

2. Materials and methods

2.1. Reagents

Sodium chloride (≥99.0%, ACS, catalog number S9888), sodium carboxymethyl cellulose (CMC, Mw ~90,000 g mol⁻¹, catalog number 419,273), and guar gum (catalog number G4129) were all purchased from Sigma-Aldrich (St. Louis, MO, USA). Calcium hydroxide (Powder, Food Codex, catalog number C1116) was acquired from Spectrum (Spectrum Chemical Mfg. Corp., New Brunswick, NJ, USA).

2.2. Alternative flour production

One kg of sweet potato tuber, peach palm fruits, and cassava roots was locally purchased, their epidermis or outer skin peeled, and the fruits’ seeds removed. After that, the totality of the pulp was freeze-dried (4.5 Plus FreeZone, ®Labconco™ Corporation, Kansas City, MO, USA). The finished flour was obtained by sieving the tissue at 0.25 mm using an ultracentrifuge mill (ZM 200 Retsch®, Haan, Germany).

2.3. Raw corn meal and nixtamalization

Commercial corn was sieved in a laboratory ultracentrifuge mill using a 500 µm sieve (Retsch ZM 200, Haan, Germany). A raw corn meal ranged from 149 to 177 microns; 96.8% of the particles (OMA™ AOAC 973.03, 2019). Afterward, 10 kg of the previously sieved corn meal were mixed with 30 L distilled water and Ca(OH)₂, and the mixture was boiled for 40 min. The cooked corn was steeped for 12 h at 22 °C. The residual liquid (nejayote, a biopolymer, López-Maldonado et al., 2017) was discarded and the cooked corn (nixtamal) was washed three times in order to remove the excess lime and pericarp (Martínez-Flores et al., 2002). The nixtamal was grounded in a mill until masa with a moisture content of ca. 50% was obtained (OMA™ AOAC 925.09, 2019). Masa with 0, 0.5, and 0.25 g lime/100 g were also prepared similarly. The fresh mixture was processed to produce tortillas, and each treatment was evaluated using physical and sensory parameters. Once the desired lime treatment was assessed, said proportions were used for other experiments (Table 1).

3. Tortilla production

All mixtures were prepared from a sample of 500 g of dry dough with and without modifications. Said modified mixtures were prepared using sweet potato, peach palm, or cassava flours. Degrees
of corn substitution in the blends were performed at two levels (i.e., 10 and 25 g each alternative flour/100 g corn mixture). Salt, CMC, and guar gum were added to the mix at 2, 0.25, and 0.25 g/100 g, respectively. Levels of gums used previously demonstrated to improve the functional and rheological properties in tortillas (Aguirre-Cruz et al., 2005; Platt-Lucero et al., 2010). Homogeneity of the blend was achieved by rotating all components by milling using a sieve of 0.25 mm. To this dough, enough water was added to obtain an ideal dough for the formation of tortillas. The hydrated dough was distributed in 50.0 g spheres; each sphere was crushed between two metal surfaces covered with plastic to form a disc of ca. 1.00 mm thick and 10.00 cm in diameter using a cast iron press (6.5 Inch, Victoria Cookware, Miami, FL, USA). Each tortilla was cooked on a hot plate at 250–270 °C for 30 s on each side. Afterward, they were left to set and cool to room temperature and stored in polyethylene bags with an airtight seal (Figure 1). Four batches of tortillas were produced (one used explicitly for sensory analysis).

3.1. Physicochemical analysis of tortilla and flours
Tortilla diameter and thickness were measured using a digital caliper (RS PRO 150 × 0.01 mm; RS Components Pte Ltd, Singapore). Assays used for flours’ characterization, such as protein, ash, potassium, sodium and calcium, zinc and copper, fat, moisture, and dietary fiber (soluble and insoluble), were performed according to OMA AOAC methods 992.23, 923.03, 985.35, 999.11, 920.85, 925.09, and 985.29/991.43, respectively. Protein, fiber, and minerals profile analysis were modified as they were assessed using a Rapid N Exceed (Elementar Analysensysteme GmbH, Langenselbold, Germany), an ANKOM TDF Fiber analyzer (ANKOM Technology, Macedon, NY, USA, Cortés-Herrera et al., 2021), and a Microwave Plasma-Atomic Emission Spectrometer (MP-AES 4210, Agilent Technologies, Santa Clara, CA, USA, Karlsson et al., 2015), respectively. Two techniques were applied to determine amino acid (Leiva et al., 2019) and fatty acid profiling (Leiva & Granados-Chinchilla, 2020). Carotenoid analysis in the flours was performed as described by Rojas-Garbanzo et al. (2016). Moisture/loss on drying was determined for tortillas immediately after preparation and two weeks after cold storage at 4–6 °C in a sealed polyethylene bag (see, section 2.9). OMA AOAC methods 996.11 and 2002.02 assessed total and resistant starch in the tortillas (Artavia et al., 2020). Finally, thermogravimetric analysis was performed using a TGA801 (LECO Corporation, Billund, Denmark, Clubbs et al., 2008; Cortés-Herrera et al., 2021). All determinations were performed in triplicate.

Table 1. Physicochemical evaluation of nixtamalized corn tortillas

| Component                  | 0       | 0.25    | 0.5     |
|----------------------------|---------|---------|---------|
| Ca(OH)₂, g/100 g           | 53.09   | 56.34   | 57.94   |
| Moisture/Loss on drying, g/100 g | 5.71    | 6.17    | 6.52    |
| Nejyote pH                  |         |         |         |

Figure 1. Comparison of tortilla mixtures. A. without substitution (negative control) and obtained by partial substitution or addition to the nixtamalized corn flour of B. 10 and C. 25 g sweet potato/100 g, D. 10 and E. 25 g/100 g peach palm and F. 10 and G. 25 g/100 g cassava flours.
3.2. Capacity of rollability
After the product cooking step, the tortilla was let to cool down until reaching 25 °C. Each specimen was manually twirled. A system of points based on a hedonic scale was established to quantitate the comportment and observations during rolling: The structure i. Cannot be rolled (1 point) ii. Collapses or breaks on both sides (2 points) iii. Collapses and breaks on only one side (3 points) iv. Cracks without breaking (4 points) v. Does not crack or break (5 points; Table 2). All determinations were performed in triplicate and assessed by the consumer panel.

3.3. Extensibility and firmness
The maximum extensibility force was determined using the method described by Cortés-Gómez et al. (2005) using a texture analyzer (TA.XTplusC, Stable Micro Systems, Surrey, United Kingdom). The compression force of a three-tier tortilla block was assessed using a spherical rounded-end probe (TA-8, ¼” diameter stainless steel ball, Texture Technologies Corp., Hamilton, MA, USA) at a speed and depth of 2 mm s⁻¹ and 30 mm, respectively. Five tortillas from each treatment were evaluated at 25 °C. The tension and compression forces are expressed as the maximum forces (g force) needed to break or cut each sample.

3.4. Swelling capacity and sensory analysis (acceptance and preference tests)
Surface puffing during sweltering, taste, texture, appearance, and color was used as a scale for sensory evaluation (Table 3). An untrained panel evaluated the physical and sensory characteristics of the tortillas where n = 32 panelists were invited, including people aged between 23–45 years and of both sexes. A subjective rating scale was used. Freshly made tortillas were evaluated, taking as reference those made with corn flour without amendment (Table 4). Sensory analyses were performed following the Guidelines for Ethical and Professional Practices for the Sensory Analysis of Foods (Institute of Food Science & Technology, 2021).

| Table 2. Parameters for tortilla rollability assessment of nixtamalized corn tortillas |
|-----------------------------------------------|---------------------------------|---------------------------------|---------------------------------|
| Physical parameters                           | It cannot be twisted            | Moderate twist                  | It cannot be twisted            |
| Capacity of rollability                        | Collapses and breaks on both sides | Cracks without collapsing       | It collapses and breaks on one side |
| Collapse                                      | None                            | 90% of the surface              | 70% of the surface              |
| Puffing                                       | Irregular                       | Irregular                       | Irregular                       |
| Rim                                           |                                  |                                 |                                 |

| Table 3. Sensory assessment of nixtamalized corn tortillas |
|------------------------------------------------------------|
| Scale for sensory evaluation of tortillas with or without substitutions |
| Assigned points | Puffing | Taste | Texture | Appearance | Color |
|-----------------|---------|-------|---------|------------|-------|
| 1               | None    | Dislike | Smooth   | Unpleasant | Unpleasant |
| 2               | Partial | Dislike slightly | Uneven | Neither | Neither |
| 3               | On all the surface | Like slightly | Rough | Pleasant | Pleasant |
| 4               | Like    |        |         |            |        |

| Sensory parameters | |
|--------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|------------------|
| Taste              | Dislike slightly (2) | Like (4) | Dislike (1) | |
| Texture            | Rough (1) | Smooth (3) | Uneven (2) | |
| Appearance         | Unpleasant (1) | Unpleasant (1) | Unpleasant (1) | |
| Color              | Neither (2) | Neither (2) | Neither (2) | |

| Total score | 6 | 10 | 7 |
Table 4. Dimensions and capacity of puffing in tortillas according to the type of cornmeal substitution

| Tortilla  | Corn | Sweet potato | Peach palm | Cassava |
|-----------|------|--------------|------------|---------|
| Addition  | 100  | 10           | 25         | 10      |
| Diameter, cm | 10.70 ± 0.37 | 10.54 ± 0.22 | 10.92 ± 0.13 | 11.06 ± 0.15 |
| Thickness, cm | 0.22 ± 0.03 | 0.27 ± 0.03 | 0.30 ± 0.07 | 0.31 ± 0.05 |
| Puffing   | 10/10 | 10/10        | 10/10      | 10/10   |

*On these scales, a grade of ten implies the formation of a puff on the totality of the tortilla surface, and zero none formed.*
3.5. Shelf life
The overall appearance of the tortilla, the sensory properties described in the previous section, and the rolling capacity at 2, 4, 7, and 14 days of storage at 4–6 °C were evaluated. Before the evaluation, the tortillas were heated in a microwave for 1 min and cooled at 25 °C. The assessment was carried out in triplicate in each treatment.

3.6. Accelerated oxidation test by OXITEST®
Thirty grams of ground tortilla were placed in the appropriate oxidation reactors in OXITEST® (Velp Scientifica, Usmate Velate, Monza and Brianza Province, Italy); the analysis was carried out from room temperature up to 90 °C, and the chambers were pressurized with oxygen according to AOCS (2017) method Cd 12c-16 (six bar with UHP gas, Praxair Technology, Inc., Uruca, San José, Costa Rica). The curves were monitored for a total of 70 h. The drop in oxygen pressure inside the oxidation chambers was constantly observed according to the ability of each treatment to oxidize. Oxisoft® (version 4.2.0, Velp Scientifica) was used to determine each induction period (IP, which is theoretically defined as the time required for a continuous oxidation cycle in the oxidation process) and a calculated shelf life using Least Mean Square Method based on each oxidation curve (Comandini et al., 2009). The shelf-life prediction was performed by measuring curves for each treatment at 20, 25, and 30 °C. The analysis was conducted twice for each sample. Each measurement was performed using two chambers where control and treatment are tested in parallel.

3.7. Statistical analysis
A one-way factorial experimental design was used, where the effect of partial substitution of cornmeal with cassava, sweet potato, and peach palm flour at two levels of 10 and 25 g/100 g was evaluated, along with the addition of 0.5 g/100 g of CMC-guar gum (both hydrogels in equal proportion). All data were analyzed by comparison of means (variance analysis) and Dunnett’s Post-Hoc method with a 95% confidence level, using the JMP® version 8 program created by Statistical Analysis System (SAS).

4. Results and discussion

4.1. Preliminary sensory evaluation of nixtamalized corn tortillas
Before any substitutions were made in the formulation of corn tortilla, lime and moisture content and pH were adjusted according to sensory and rheological parameters. Three different lime and moisture concentrations were tested, and 0.25 and 56.34 g/100 g were selected as the best parameters to formulate (Table 1). Under these conditions, the wastewater pH is not very high, resulting in a nejayote pH of 6.17. These pH values align with what Mexican legislation suggests (i.e., 5.5–10). Nejayote has a negative surface charge throughout the pH range (López-Maldonado et al., 2017). Selected moisture and calcium values align with those reported elsewhere (Clubbs et al., 2008; Valderrama-Bravo et al., 2010). Clubbs et al. (2008) reported ca. 8 g/100 g moisture loss during the cooking process.

4.2. Physicochemical analysis of the flours used for substitution

4.2.1. Amino acid profiling in the flours
Amino acid analysis shows that the substituents’ profiles are somewhat similar, with a few exceptions (Table 5). Overall, the cassava flour profile exhibits lower values in amino acids (Table 5). Contrary to cassava flour, sweet potato and peach palm flours will maintain histidine values instead of diminishing them. Arginine and phenylalanine are lowered in all cases but are more noticeable in cassava substitution cases. Interestingly, corn nixtamalization improves histidine profile (470.6 ± 23.8 vs. 1799.1 ± 90.9 mg/100 g), decreases values of Arg (2408.0 ± 121.7 vs. 1066.5 ± 53.9 mg/100 g), and has no effect on glutamic acid. In this scenario, the objective of the substitution should be maintaining the levels of amino acids in corn and nixtamalized flour in cases where augmenting them is not possible. Corn is usually considered a deficient food in lysine and tryptophan (Mahan et al., 2014).
Table 5. Physicochemical characterization of flours used as raw ingredients for tortilla production

| Ingredient                                                                 | Sweet potato | Peach palm | Cassava | Corn | Nixtamalized corn |
|----------------------------------------------------------------------------|--------------|------------|---------|------|-------------------|
| **Protein and amino acid profile**                                         |              |            |         |      |                   |
| Protein (Nitrogen), g/100 g                                                | 3.89 ± 0.09 [0.62 ± 0.01] | 6.49 ± 0.16 [1.04 ± 0.02] | 1.45 ± 0.03 [0.23 ± 0.01] | 7.61 ± 0.18 [1.22 ± 0.03] | 8.03 ± 0.19 [1.29 ± 0.03] |
| Asparagine, mg/100 g                                                      | 644.3 ± 32.6 | 904.1 ± 45.7 | 163.3 ± 8.3 | 337.7 ± 17.1 | 516.4 ± 26.1 |
| Glutamine, mg/100 g                                                       | 372.3 ± 18.8 | 562.6 ± 28.4 | 238.9 ± 12.1 | 1389.4 ± 70.2 | 1319.7 ± 66.7 |
| Serine, mg/100 g                                                          | 460.4 ± 23.3 | 700.1 ± 35.4 | 84.4 ± 4.3 | 207.8 ± 10.5 | 887.1 ± 44.8 |
| Histidine, mg/100 g                                                       | 1 055.7 ± 53.3 | 1 703.3 ± 86.1 | 161.8 ± 8.2 | 470.6 ± 23.8 | 1799.1 ± 90.9 |
| Glycine, mg/100 g                                                         | 196.0 ± 9.9 | 324.8 ± 16.4 | 61.0 ± 3.1 | 285.2 ± 14.4 | 315.6 ± 15.9 |
| Threonine, mg/100 g                                                       | 99.6 ± 5.0 | 376.4 ± 19.0 | 112.2 ± 5.7 | 487.6 ± 24.6 | 424.0 ± 21.4 |
| Arginine, mg/100 g                                                        | 374.5 ± 18.9 | 568.7 ± 28.7 | 215.9 ± 10.9 | 2 408.0 ± 121.7 | 1 066.5 ± 53.9 |
| Alanine, mg/100 g                                                         | 43.4 ± 2.2 | 92.2 ± 4.7 | 129.5 ± 6.5 | 86.9 ± 4.4 | 97.9 ± 4.9 |
| Tyrosine, mg/100 g                                                        | 82.8 ± 4.2 | 239.0 ± 12.1 | 15.7 ± 0.8 | 57.9 ± 2.9 | 162.9 ± 8.2 |
| Cysteine/Cystine, mg/100 g                                                | 155.4 ± 7.9 | 296.4 ± 15.0 | 70.2 ± 3.5 | 354.3 ± 17.9 | 287.8 ± 14.5 |
| Valine, mg/100 g                                                          | 84.9 ± 4.3 | 118.7 ± 6.0 | 38.0 ± 1.9 | 76.1 ± 3.8 | 138.2 ± 7.0 |
| Methionine, mg/100 g                                                      | 74.1 ± 3.7 | 144.3 ± 7.3 | 58.9 ± 3.0 | 205.5 ± 10.4 | 145.5 ± 7.4 |
| Phenylalanine, mg/100 g                                                   | 153.9 ± 7.8 | 264.9 ± 13.4 | 35.8 ± 1.8 | 725.2 ± 36.6 | 613.8 ± 31.0 |
| Isoleucine, mg/100 g                                                      | 26.5 ± 1.3 | 64.0 ± 3.2 | 23.2 ± 1.2 | 253.5 ± 12.8 | 80.8 ± 4.1 |
| Leucine, mg/100 g                                                         | 32.2 ± 1.6 | 32.4 ± 1.6 | 30.4 ± 1.5 | 163.4 ± 8.3 | 82.1 ± 4.1 |
| Lysine, mg/100 g                                                          | 34.2 ± 1.7 | 98.0 ± 5.0 | 11.3 ± 0.6 | 100.9 ± 5.1 | 92.7 ± 4.7 |
| **Mineral profile**                                                        |              |            |         |      |                   |
| Ash, g/100 g                                                              | 2.67 ± 0.13 | 2.77 ± 0.14 | 1.64 ± 0.08 | 1.26 ± 0.06 | 2.72 ± 0.14 |
| Potassium, g/100 g                                                        | 0.81 ± 0.05 | 0.47 ± 0.03 | 0.37 ± 0.02 | 0.27 ± 0.02 | 0.30 ± 0.02 |
| Sodium, mg kg⁻¹                                                           | 1 118.88 ± 111.89 | 7 350.43 ± 735.04 | 489.51 ± 48.95 | 349.65 ± 34.92 | 260.3 ± 26.03 |
| Calcium, mg kg⁻³                                                          | 880.41 ± 112.99 | 867.30 ± 111.31 | 536.29 ± 68.83 | 246.19 ± 31.60 | 900.39 ± 115.56 |
| Zinc, mg kg⁻¹                                                             | 6.17 ± 1.25 | 7.15 ± 1.45 | 5.05 ± 1.02 | 15.46 ± 3.14 | 164.2 ± 3.33 |
| Copper, mg kg⁻³                                                           | 5.38 ± 1.17 | 4.27 ± 0.92 | 1.46 ± 0.32 | 1.59 ± 0.34 | 1.28 ± 0.28 |

(Continued)
| Ingredient          | Sweet potato | Peach palm | Cassava | Corn      | Nixtamalized corn |
|---------------------|-------------|------------|---------|-----------|-------------------|
| **Fat and fatty acid profile, g/100 g** |             |            |         |           |                   |
| Crude fat           | 1.07 ± 0.01 | 9.37 ± 0.11| 0.74 ± 0.01| 4.79 ± 0.06| 4.72 ± 0.06       |
| C10:0               | 9.90 ± 0.12 |           |         |           |                   |
| C12:0               | 3.07 ± 0.04 |           |         |           |                   |
| C14:0               |             | 0.10 ± 0.01|         |           |                   |
| C16:0               |             | 0.53 ± 0.01|         |           | 18.91 ± 0.22      |
| 9c11t C18:2         |             | 6.78 ± 0.08| 0.21 ± 0.02| 0.62 ± 0.01|                   |
| 10c12t C18:2        |             | 36.73 ± 0.43| 0.25 ± 0.03| 17.46 ± 0.20|                   |
| C18:0               | 2.26 ± 0.03 | 6.31 ± 0.07| 0.76 ± 0.01| 34.84 ± 0.51| 33.72 ± 0.40      |
| H2NCOOH             |             | 5.75 ± 0.07| 55.83 ± 0.67|           |                   |
| 9c C18:1            | 0.34 ± 0.01 | 41.21 ± 0.58| 42.98 ± 0.61| 25.68 ± 0.30| 26.58 ± 0.32      |
| 9c C16:1            |             | 1.23 ± 0.12|         |           |                   |
| 9c12c C18:2         | 31.34 ± 0.37| 1.24 ± 0.09|         | 16.59 ± 0.20| 19.22 ± 0.25      |
| 9c15c C18:2         |             |             |         |           | 0.14 ± 0.02       |
| 11c14c17c C20:3     | 0.16 ± 0.01 |             |         |           | 0.34 ± 0.04       |
| 2,4,4-Me C6:0       | 0.42 ± 0.03 | 0.14 ± 0.05|         |           |                   |
| 2-Me C4:0           | 50.11 ± 0.59|             |         |           |                   |
| 1,4-C6H4(COOH)2     |             |             |         | 3.61 ± 0.34| 2.96 ± 0.03       |
| Σ SFA               | 65.76       | 12.59       | 56.73   | 57.46     | 36.68             |
| Σ MUFA              | 0.34        | 42.44       | 42.98   | 25.68     | 26.58             |
| Σ PUFA              | 31.34       | 44.91       | 0.00    | 17.53     | 37.30             |

**Ethanol-soluble sugars, g/100 g dry weight basis**

| Ingredient | Sweet potato | Peach palm | Cassava | Corn | Nixtamalized corn |
|------------|-------------|------------|---------|------|-------------------|
| Fructose   | 1.86 ± 0.26 | < 0.19     | < 0.19  | < 0.19| < 0.19            |
| Glucose    | 2.75 ± 0.43 | < 0.22     | < 0.22  | < 0.22| < 0.22            |
| Sucrose    | 7.51 ± 0.88 | < 0.72     | 1.27 ± 0.15| 1.51 ± 0.18| < 0.72        |

(Continued)
| Ingredient | Sweet potato | Peach palm | Cassava | Corn   | Nixtamalized corn |
|------------|--------------|------------|---------|--------|------------------|
| Dietary Fiber | 7.82 ± 1.37  | 4.56 ± 0.80 | 1.91 ± 0.21 | 8.21 ± 1.44 | 8.65 ± 1.52 |
| Soluble    | 3.85 ± 0.67  | 0.27 ± 0.05 | 0.72 ± 0.13 | 1.42 ± 0.25 | 2.02 ± 0.35 |
| Insoluble  | 3.97 ± 0.70  | 4.29 ± 0.75 | 1.19 ± 0.21 | 6.79 ± 1.19 | 6.56 ± 1.15 |
| Other assays, g/100 g | | | | | |
| Moisture   | 9.20 ± 0.64  | 8.51 ± 0.60 | 7.80 ± 0.55 | 13.60 ± 0.95 | 10.80 ± 0.76 |
4.2.2. Mineral profiling in the flours
Regarding mineral profile, [Ca\(^{2+}\)] was not affected by the partial substitution of corn with sweet potato and peach palm; a contrary effect was observed in the case of cassava flour (Table 5). A relevant feature is that calcium absorption from corn tortillas is relatively high (Rosado et al., 2005). When compared to the nixtamalized flour, [Zn\(^{2+}\)] concentration decreased in all treatments (Table 5). Sweet potato and peach palm flours tend to increase considerably [Na\(^{+}\)] (the latter substantially more than the former) within the final tortilla formulation (Table 5). An effect may not be desirable since consumer products continuously shift toward a low sodium intake (Hyseni et al., 2007). Finally, sweet potato flour inclusion also increases [K\(^{+}\)] ion (Table 5). Usually, roots and tubers crops are deficient in most other vitamins and minerals. Still, sweet potato tends to retain relatively high levels of Na and K. Like other crops, the nutritional value of roots and tubers varies with variety, location, soil type, and agricultural practices, among others (Chandrasekara & Kumar, 2016).

4.2.3. Fatty acid profiling in the flours
In the case of fat input, the peach palm will incorporate almost double the fat content that corn contributes (i.e., 9.37 ± 0.11 vs. 4.72 ± 0.06 g/100 g, Table 3). While Zea mays is a Poaceae, palms from the Arecaceae family are commercially valuable for their fruit capacity to produce oil (Correa Martins et al., 2014). However, the fat profile in the peach palm is highly desirable as fat is mainly mono and polyunsaturated (42.44 and 46.14 g/100 g) and has a low saturated to polyunsaturated fatty acid ratio (0.15), and is not modified during cooking (Fernández-Piedra et al., 1994). For example, peach palm flour’s relatively higher fat content could explain increased tortilla softness and pliability by reducing the rate of staling observed (Clubbs et al., 2008).

4.2.4. Sugars profiling in the flours
As expected, the main difference among the formulations used is the input of sugars that the sweet potato flour will introduce to the final tortilla formulation. These compounds will impact sensory aspects, including the tortilla’s color, taste, and texture, via the Maillard reaction during cooking (Liska et al., 2016). Though cooking sweet potato has been demonstrated to favor health injuring compounds such as hydroxymethylfurfural and acrylamide (Truong et al., 2014), nixtamalization has also been shown to reduce levels of such substances (Topete-Betancourt et al., 2019). The amount of sugars present in the formulation with sweet potato may be why it was the least preferred tortilla by the panelists (Table 5, see below).

4.2.5. Dietary fiber in the flours
Dietary fiber, at least for corn flour, is in line with those reported earlier (i.e., 7.7–12.0 g/100 g; Bressani et al., 2001). Finally, dietary fiber content was not affected by the inclusion of sweet potato flour as the value was very similar to both corn and nixtamalized corn flours (8.21 ± 1.44 and 8.65 ± 1.52 g/100 g, respectively). (Table 5). The inclusion of the other two flours decreases this value considerably, as their input is comparably low (4.56 ± 0.80 and 1.91 ± 0.20 g/100 g, Table 5). Again, an undesirable effect considering dietary fiber health benefits (Anderson et al., 2009).

4.2.6. Carotenoid input from flours to tortillas
Carotenoid content obtained for the flours varied, in decreasing order, from 205 to 98, 56 to 42, and 9 to 7 μg β-carotene g\(^{-1}\) dry weight basis for peach palm, sweet potato, and cassava flours, respectively. These values align with those previously described (Atwijukire et al., 2019; Rodriguez-Amaya et al., 2011). In the case of sweet potato and cassava flours, our data sets the variety as light-yellow clones (Atwijukire et al., 2019) and yellow to deep orange-fleshed varietals (Rodriguez-Amaya et al., 2011), respectively. There is an inherent advantage in using carotenoid-containing flours as enrichment in nixtamalized corn (especially considering that it is a relatively poor source of β-carotene; Rodriguez-Amaya et al., 2011). In this case, carotenoids impart, among other properties, nutraceutical properties to the product (Arguedas-Gamboa et al., 2015). All flours incorporate carotenoids into the processed product (Alano Vieira et al., 2018; Joeger de Carvalho et al., 2017; Jatunov et al., 2010; Rosales et al., 2016; Vimala et al., 2011). Retention of these compounds in tortillas has already been demonstrated
(Arguedas-Gamboa et al., 2015; Rosales et al., 2016). Finally, in the particular case of carotenoids, the nixtamalization process seems to improve their bioavailability (Colín-Chávez et al., 2020). Though some consumers prefer white corn for food consumption, pigmented corn has gained considerable interest (Colín-Chávez et al., 2020). Color-containing flours can indeed impact sensory see, for example, the score for color and appearance observed for sweet potato, despite being proficient in other qualities.

4.3. Physical characteristics of the substituted tortillas

4.3.1. Moisture and puffing in a tortilla

The typical puffing for a traditional tortilla is observed only with sweet potato substitutions and the cassava substitution at a 10 g/100 g level (Table 4). Puff during tortilla cooking process has been considered a subjective test (Figueroa et al., 2001; Treviño-Mejía et al., 2016). Flour moisture content plays a significant role in tortilla puffing during cooking (Table 5). As heat is applied, the moisture is lost puffing occurs on the surface of the tortilla. This characteristic is relevant as it indicates adequate cooking (Reyes-Moreno et al., 2013). Hence, tortilla surface puffing is considered part of the sensory descriptive analysis (Winger et al., 2014).

4.3.2. Moisture behavior during tortilla storage

Tortillas with both substitution levels with sweet potato showed significant differences with respect to the control (p < 0.05) for days 1 and 15. In this regard, sugars and carbohydrates (e.g., starch) can influence moisture uptake (Table 5; Browne, 1922). Meanwhile, cassava formulations showed significant differences at day 15 (unlike other formulations, a decrease in moisture was observed; Figure 2A). Cassava flour has a higher starch content compared to the other flours (i.e., 50.57, 54.93, 83.37, 58.80 g/100 g for corn, sweet potato, peach palm, and cassava, respectively; Artavia et al., 2020), and probably with a high proportion of amylose, which increases water loss and retrogradation (see below). Hydrocolloids, used traditionally as thickening and gelling agents, have also been described as cryoprotectants (Maity et al., 2018; Saha & Bhattacharya, 2010). Finally, it has been previously described that the traditional tortilla formulation usually retains 50–60 g/100 g of water. These values are in line with those reported elsewhere (Clubbs et al., 2008). Also, moisture content can be related to water activity (as high as 0.98) and, in turn, to thermal behavior and shelf life (Clubbs et al., 2008; Tellez-Giron et al., 1988).

4.3.3. Capacity of rollability during tortilla storage

There was a significant difference (p < 0.05) in the quality of rolling for the 25 g/100 g substitution using peach palm and cassava flour (Table 6; Figure 2B). The tortilla cannot be easily rolled in both cases and exhibits a break on both sides. Additionally, in these formulations, a smaller volume of water was absorbed for the dough formation; therefore, it can be considered that an adequate gelling of the starch present was not complete.

The behavior observed regarding extensibility/firmness and rollability are in line with those reported elsewhere, where they trend inversely as time progresses (Clubbs et al., 2008; Mao & Flores, 2001). This tendency is especially seen in wheat tortilla composites (Asiyanbi-Hammed & Simsek, 2020). However, some mechanical properties are improved for wheat due to higher gluten and protein content (Diztek et al., 2022). On the other hand, Platt-Lucero et al. (2012) already observed an increase in extensibility and a decrease in rollability in just 48 h in tortillas prepared with xanthan gum. In this regard, the mixture used herein imports better mechanical characteristics.

Fresh tortillas present a greater softness and are easily rolled without breaking. As time passes, the rigidity increases, and the rolling capacity is diminished due to the loss of water by retrogradation of the gel that makes up the tortilla (Suhendro et al., 1998). Throughout the storage, there was a decrease in the capacity of tortilla roll in the three formulations (Table 6; Figure 2B). The substitutions with sweet potato flour showed higher resistance to losing the ability to roll (i.e., maintained over time). Again, flour with a more significant amount of carbohydrates and,
consequently, a higher capacity for water retention. The reverse is also true for the substitution with cassava; water loss occurs at a higher rate as it has a lower amount of simple carbohydrates to retain (Table 5, Figure 2A).

After 15 days of storage, the rolling capacity of most of the tortillas was lost, and microbiological development was perceived. Although sweet potato-supplemented tortillas retain their ability to roll up from the 7th day of storage, an unpleasant odor and mold formation are perceived on the surface, representing the formulation with the shortest shelf life.

Table 6. Rheological analysis of the different products made with substitution of cornmeal for non-traditional flours

| Treatment                  | Days of storage | Capacity of rollability |
|----------------------------|-----------------|-------------------------|
|                            | 0   | 2    | 4    | 7    | 15   |
| Control                    | 4.67| 3.67 | 3.67 | 3.33 | 2.00a|
| Sweet potato 10 g/100 g    | 5.00| 4.33 | 5.00 | 4.33 | 4.33 |
| Sweet potato 25 g/100 g    | 4.00| 4.67 | 4.67 | 4.00 | 4.33 |
| Peach palm 10 g/100 g      | 3.67| 5.00 | 4.00 | 3.33 | 3.00 |
| Peach palm 25 g/100 g      | 2.00| 3.33 | 2.67 | 2.67 | 1.67 |
| Cassava 10 g/100 g         | 4.67| 4.67 | 4.33 | 4.00 | 2.00a|
| Cassava 25 g/100 g         | 2.67| 2.67 | 2.67 | 3.00 | 2.00 |

*Values differ significantly from the control (p < 0.05) based on the post hoc Dunnet test.
4.4. Other physicochemical characteristics of the substituted tortillas

4.4.1. Total (TS) and resistant starch (RS) in the tortilla

Total starch for the substituted tortillas sweet potato, peach palm, cassava, and control, were 60.0 ± 3.1, 58.8 ± 3.1, 87.8 ± 4.6, and 85.30 ± 4.5 g/100 g, respectively. Meanwhile, values of RS of 0.945 to 1.336 (cassava > peach palm > sweet potato) and 0.814 g/100 g were obtained for the substituted flours and control (nixtamalized corn flour), respectively (Figure 2C). In all cases, the amount of RS increased as the degree of substitution increased (Figure 2C). Each starch source has different characteristics according to its origin; starch enzymatic digestibility evaluates dietary nutritive input (Artavia et al., 2020). For example, cassava starch resists the least to enzymatic breakdown (using extracellular or microbial amylases). On the other hand, sweet potato starch was more susceptible than cassava starch to degradation by a-amylase and glucoamylase (Moorthy, 2002). Our data exhibit lower RS values than those reported elsewhere for resistant starch (i.e., 2.49 to 5.23 g/100 g) in tortillas with hydrocolloids (Rendón-Villalobos, Agama-Acevedo, Islas-Hernández, Paredes-López et al., 2006a). However, they compare more closely to retrograded RS (i.e., 1.23 to 1.76 g/100 g) reported in the study mentioned above (Rendón-Villalobos, Agama-Acevedo, Islas-Hernández, Paredes-López et al., 2006a). Interestingly, values were reported by Enríquez-Castro and coworkers (2020) for extruded and traditional nixtamalization processes (0.79 to 1.01 g/100 g). Furthermore, Rojas-Molina et al. (2020) recently reported three types of RS (inaccessible starch [RS1], retrograded starch [RS3], and V-amylase-lipid complexes [RS5]). They demonstrated that the RS increases for corn-based tortillas within 30 days at 4 °C storage. Hence, we expect the substituted flours to exhibit comparatively more RS during refrigeration.

4.4.2. Shear force and deformation analysis of freshly made tortillas and behavior during storage

Objective texture determinations were assessed. Overall, the partial substitution of corn flour improved the resistance to being cut and deformed (Table 7). Substitution with sweet potato increase resistance due to its capacity to absorb water; the gel formed will have a better consistency and greater firmness (Hernández-Medina et al., 2008). Throughout storage, the cutting force increased in all cases, a trend reported previously for other tortillas (Mao & Flores, 2001). For example, the force required to puncture corn tortillas has been estimated at 0.12–0.15 N and, as seen here as well, increases with storage time as moisture decreases (Román-Brito et al., 2007). The substitution with palm peach flour caused the cutting force to grow more rapidly than in the other substitutions (Table 7). The contrary is observed for the sweet potato formulations (Table 7). On the other hand, the compression force (i.e., the difference in the elasticity of the tortilla) showed no significant difference (p > 0.05) among substitutions (Table 7).

4.4.3. Accelerated oxidation and predicted shelf life of tortillas

In line with the sensory analysis, induction periods for the control were significantly lower (p < 0.05) than those for sweet potato (Table 8). No differences were observed between the control and cassava substituted tortillas. Finally, substituted flours with addition higher than 10 g/100 g seem to have no effect over induction periods (Table 8), which indicates that several advantageous properties of the non-traditional flours can be achieved with relatively small proportions. Interestingly, OXITEST® has been used for other baked products (especially taralli; Veranto et al., 2010), but no accelerated oxidation has been reported for tortillas (Artavia et al., 2021). Traditional methods (including microbial degradation) for determining the shelf life of tortilla and other corn dough products have been reported previously (Ordaz Ortiz & Vázquez Carrillo, 1997; Aida et al., 1996; Martínez-Flores et al., 2004; Hernández Montaya et al., 2020). In line with our results (see, Table 6), tortilla shelf life has been reported from 3 to 6, and from 8 to 16 days at 22 and 4 ± 1 °C; the lower values correspond to tortillas without the addition of preservatives (Martínez-Flores et al., 2004). Additives and low temperature can improve shelf life up to 8-fold (Martínez-Flores et al., 2004; Ordaz Ortiz & Vázquez Carrillo, 1997). Finally, the results indicate that
Overall, the tortillas with 25 g/100 g sweet potato flour have a significantly lower rating compared to the control (p < 0.05), whilst the other formulations do not differ (Table 9). The products were also assessed according to their texture, general appearance, color, and taste or flavor; the collected results are summarized in Table 9. Regarding the surface, no significant differences were detected (p > 0.05). A considerable rejection was perceived in the appearance of the tortilla with 25 g/100 g sweet potato, which is related to the color of tortillas (Figure 1). As for the flavor, a rejection is prompted with the tortillas with 25 g/100 g peach palm flour.

### 4.5. Sensory analysis of tortillas

Overall, the tortillas with 25 g/100 g sweet potato flour have a significantly lower rating compared to the control (p < 0.05), whilst the other formulations do not differ (Table 9).
### Table 9. Sensory analysis of the different products made with substitution of cornmeal for non-traditional flours, evaluated by $n = 32$ panelists

**General Acceptance**

| p-value       | Treatment               | Averages$^b$ | Dunnet p-value |
|---------------|-------------------------|--------------|----------------|
| 0.0071        | Control                 | 87.63        | 1              |
| Levene test   | Sweet potato 10 g/100 g | 77.26        | 0.4625         |
| 0.0134        | Sweet potato 25 g/100 g | 68.99$^a$    | 0.0147         |
| Shapiro-Wilk  | Peach palm 10 g/100 g  | 84.31        | 0.9233         |
| 0.0049        | Peach palm 25 g/100 g  | 77.75        | 0.2872         |
|               | Cassava 10 g/100 g     | 93.25        | 0.9386         |
|               | Cassava 25 g/100 g     | 81.21        | 0.9873         |

**Treatment**

| Evaluated aspects | Taste | Texture | Appearance | Color |
|-------------------|-------|---------|------------|-------|
| Control           | 2.7   | 2.5     | 2.3        | 2.5$^a$ |
| Sweet potato 10 g/100 g | 2.4   | 2.7     | 2.1$^a$    | 2.1$^a$ |
| Sweet potato 25 g/100 g | 3.1   | 2.3     | 2.9        | 3.0   |
| Peach palm 10 g/100 g | 2.8   | 2.1     | 2.9        | 2.8   |
| Peach palm 25 g/100 g | 2.1$^a$ | 2.2   | 2.7        | 2.9   |
| Cassava 10 g/100 g | 3.2   | 2.7     | 3.0        | 3.2   |
| Cassava 25 g/100 g | 2.8   | 2.4     | 2.8        | 2.9   |

$^a$Values differ significantly from the control ($p < 0.05$) based on the post hoc Dunnet test. $^b$Overall qualifications, the panel gave each tortilla treatment on a scale from 1 to 100, and individual scores were used to calculate a mean.

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**Figure 3.** The typical TGA curve of corn tortillas shows the weight loss for A. flours used as raw material and B. tortilla treatments. The derivative weight loss curve is also offered as a function of temperature for both cases C. and D., respectively. The derivative weight loss curve was further characterized by deconvolution.
4.6. Thermogravimetric analysis of tortillas
A thermal study of the flours (Figure 3A) shows decomposition temperatures over 290 °C (corn > peach palm > cassava) correspond to the degradation of starch (X. Liu et al., 2010). In contrast, other reports have related this temperature with gluten and protein structure. However, it is more likely related to amylose/amyllopectin differences (X. Liu et al., 2010), as all of the ingredients used herein are gluten-free but share ~290 °C inflection (Figure 3A, 3B). On the other hand, sweet potato flour has a corresponding peak at 267.3 °C, significantly lower than the other flours, which can be related to their lower RS values. Some peculiarities regarding sweet potato behavior during thermal analysis have already been pointed out (Y. Liu et al., 2019).

TGA for tortillas exhibited two peaks (Figure 3C,D). Peak 1, which resembles a valley, has temperatures between 185 °C and 255 °C and increases with the flour substitution percentage. This valley can be explained as a result of the evaporation of the water occluded in the starch bags formed during the tortilla cooking process. Peak 2, corresponding to the starch decomposition, have values between 286 °C and 302 °C, with an increasing temperature when the flour substitution percentage increase, except for the sweet potato (10% = 299.8 °C and 25% = 286.2 °C). This result can be related to a lower amount of amylose, as reported by (X. Liu et al., 2010; Pineda-Gómez et al., 2011); the amylose polymer form a coiled–helical structure which is a very compact physical structure that requires more thermal energy for its decomposition (Pineda-Gómez et al., 2011). On the other hand, amyllopectin polymers form a less compact structure due to their α-1,6 bonds responsible for its ramification.

5. Conclusions
Corn nixtamalized corn can be successfully substituted with non-traditional flours. Partial substitutions with different starch sources can impart other mechanical and chemical properties to the tortilla. The incorporation of such flours in the traditional tortillas is not only possible but can impact food preservation and improve specific nutritional properties. However, the substitution should be carefully balanced to enhance the degree of acceptance by the consumer, as negative sensory perception could counterbalance benefits and hinder possible commercially viable products. Hence, in this regard, enriched corn flour may serve as a vehicle to incorporate nutraceutical-rich ingredients (e.g., carotenoids) with the potential to improve the original flour composition. Such additions or modifications can provide nutritional and sensorial diversification, especially for diets with restrictions (e.g., gluten-free diets). Additionally, the alternative flours assessed herein could include other dietary benefits such as a more favorable lipolytic profile towards the unsaturated fatty acids. From the assayed flours, sweet potato provided the most improvements within the physicochemical parameters tested and might be an exciting avenue to explore further. Other sweet potato cultivars might be used to incorporate other sensory, nutritional, and chemical properties (e.g., purple or red sweet potatoes, colors provided by anthocyanins, Ji et al., 2015).

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