Ultrasound-assisted modification and multianalytical characterization of organic *Arracacia xanthorrhiza* starch

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Keywords:

Starch
Arracacia xanthorrhiza
Ultrasound
Modification
Thermal stability

Article history:

Received 25.06.2025
Received in revised form 23.09.2025
Accepted 30.09.2025

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DOI:
10.24263/2304-974X-2025-14-3-4

Abstract

Introduction. Native starches have certain industrial limitations, making it necessary to apply physical modification methods such as ultrasound treatment. This study investigates the effects of ultrasound on the structural, thermal, and physicochemical properties of organic starch extracted from *Arracacia xanthorrhiza*.

Materials and methods. The *Arracacia xanthorrhiza* roots were obtained from a small-scale producer in Curitiba, Paraná, Brazil. Starch was extracted using an aqueous method, modified by ultrasound, and subsequently characterized in terms of thermal, pasting, morphological, structural, and physicochemical properties.

Results and discussion. The aqueous extraction of mandioquinha-salsa starch yielded 34.64% based on raw material and 48.18% on processed mass, showing values consistent with previous studies. The application of ultrasound at different amplitudes (40%, 50%, 60%, and 70%) resulted in notable structural and functional changes in the starch. Rapid Visco Analysis revealed that the 40% amplitude treatment improved shear resistance and reduced breakdown and retrogradation, while higher amplitudes increased peak and final viscosities. Thermogravimetric (TG) and derivative thermogravimetric (DTG) analyses showed that the 50% amplitude treatment resulted in the highest degradation temperature, while the 60% treatment exhibited the widest range of thermal stability. Differential Scanning Calorimetry (DSC) analysis confirmed lower gelatinization enthalpy for the 40% treated starch, indicating reduced energy demand during gelatinization. FEG-SEM (Field Emission Gun – Scanning Electron Microscope) images demonstrated changes in granule morphology, with increased particle size up to 50% amplitude, followed by a reduction at higher intensities, without evidence of granule agglomeration. X-ray diffraction showed a gradual increase in crystallinity with ultrasound treatment, although all modified samples had lower crystallinity than the native starch. These results demonstrate that ultrasound is an effective and clean modification method, capable of enhancing the thermal stability, viscosity behavior, and structural organization of starch from *Arracacia xanthorrhiza*, making it suitable for food and packaging applications.

Conclusions. Ultrasound-modified starches from *Arracacia xanthorrhiza* showed improved functional and thermal properties compared to native starch.

Introduction

Starches, like cellulose, are among the most abundant carbohydrates in nature, and their structural and functional properties vary widely depending on their botanical origin. These differences are related to the amylose-to-amylopectin ratio, granule size, and morphology, directly affecting their applications in the food, pharmaceutical, and packaging industries (Zhang et al., 2023). Industrial interest in native and functional starches has grown, particularly due to the technological properties of resistant starch, which is associated with glycemic modulation, gut health, and beneficial effects on the microbiota (Bojarczuk et al., 2022; Li, 2022).

However, native starches present technological limitations such as retrogradation, syneresis, and low thermal stability, which compromise their industrial use. To overcome these limitations, various modification methods have been applied, including chemical, enzymatic, and physical modifications (Giacomozzi et al., 2021; Wang et al., 2023).

Among physical methods, ultrasound application stands out as a clean and environmentally friendly approach, promoting structural changes in starch granules through acoustic cavitation. This technique produces granules that are more porous, with reduced particle size, altered crystallinity, and improved solubility, all without using toxic reagents or harsh processes (Zhu, 2015). Ultrasonic modification has been widely studied across various starch sources, proving effective in enhancing functional properties.

Mandioquinha-salsa (*Arracacia xanthorrhiza*) is an Andean tuber widely cultivated in Brazil, especially in the South and Southeast regions. Its starch is characterized by high digestibility, fine granular morphology, and a high amylopectin content, making it a good candidate for structural modifications aimed at greater stability and functionality.

Therefore, this study aims to evaluate the effects of ultrasonic treatment on organic mandioquinha-salsa starch using different amplitudes and exposure times. The samples were characterized by thermal analyses (DSC/TG), pasting properties (RVA), electron microscopy, and physicochemical evaluation, seeking to demonstrate the viability of this technique as a tool to generate functional starches with high technological potential.

Materials and methods

Starch extraction

Organic *Arracacia xanthorrhiza* roots (yellow common variety) were obtained from a local producer in Curitiba (Paraná, Brazil). The roots were washed, peeled, sliced into 3–5 mm pieces, and stored under refrigeration (5 °C) until use.

Starch extraction was performed using an aqueous method. Enzymatic inactivation was carried out by immersing the sliced material in a sodium metabisulfite solution ($Na_2S_2O_5$) for 30 minutes, followed by rinsing in running water.

The aqueous grinding was performed in an industrial blender using a fixed proportion of root to water. The homogenized suspension was sieved (270 mesh), centrifuged, and the resulting starch was dried in a forced-air oven at 35 °C for 24 hours. The dried starch was then manually ground with a mortar and pestle.

The native starch was stored in a desiccator containing anhydrous calcium chloride until further modification and analysis.

Starch modification

The starch granules were modified by ultrasound treatment using a Vibra-Cell 500 W device (Sonics & Materials Inc., USA) operating at a frequency of 20 kHz. A 10% (w/v) starch suspension in deionized water was subjected to ultrasonic waves while being kept in an ice bath to prevent starch gelatinization during the process. The total ultrasound exposure time was 30 minutes, as described by Liu et al. (2023).

Analysis

Thermogravimetric analysis (TG) was performed using a TGA-50 instrument (Shimadzu, Japan) under the following conditions: sample mass of approximately 7.0 mg; air atmosphere with a flow rate of 150 mL min^{-1}; heating rate of 10 °C min^{-1} from 30 °C to 650 °C. Mass losses were calculated using the TA-60 analysis software.

Differential scanning calorimetry (DSC) analyses were performed using a DSC-Q200 instrument (TA Instruments, USA), previously calibrated. The experimental conditions were as follows: air flow rate of 50 mL min^{-1}, heating range from 20 °C to 100 °C, and heating rate of 10 °C min^{-1}. Approximately 2.5 mg of each sample was weighed and mixed with 10 µL of distilled water (1:4, w/v) in aluminum pans, which were then hermetically sealed. The pans were allowed to stand for 1 hour before analysis to ensure moisture equilibrium and starch granule swelling.

X-ray diffraction (XRD) analysis was carried out using an Ultima IV diffractometer (Rigaku, Japan), located at the Multiuser Laboratory Complex (C-LABMU) of the State University of Ponta Grossa (UEPG). The measurements were performed using CuKα radiation (λ = 1.541 Å), with the equipment set to 40 kV and 20 mA. The scattered radiation was recorded over a 2θ range from 3° to 40°, with a scanning speed of 2° min^{-1} and a step size of 0.06° (Kubiaki et al., 2016).

Pasting properties were evaluated using a Rapid Visco Analyzer (RVA), model RVA-4 (Newport Scientific, Australia). Native and ultrasound-modified starch suspensions were prepared at 8% (w/w, dry basis) in deionized water, with a final mass of 28 g. The controlled heating and cooling cycle consisted of constant stirring at 50 °C for 2 minutes, followed by heating from 50 °C to 95 °C at a rate of 6 °C min^{-1}. The sample was then held at 95 °C for 5 minutes, cooled back to 50 °C at the same rate, and held at 50 °C for an additional 2 minutes, according to the method described by Balet et al. (2019).

The diameter and shape of the mandioquinha-salsa starch granules were observed using Field Emission Gun – Scanning Electron Microscope (FEG-SEM) model MIRA 3 (Tescan, Czech Republic). Samples were mounted on carbon tapes and coated with gold plasma to ensure electron conductivity. Analysis was conducted at an accelerating voltage of 20 kV with magnifications of 1000x and 2000x, as described by Cremasco (2023).

Statistical analysis

The results of the analyses were expressed as mean followed by standard deviation. Analysis of variance (ANOVA) and Tukey's test were used to compare the means between the samples with 95% confidence ($p < 0.05$).

Results and discussion

The starch extraction process from *Arracacia xanthorrhiza* roots began with 4,980 g of raw material, previously selected and cleaned to remove surface impurities, peels, and fibrous residues. After this preparation, 3,580 g of usable fresh matter were obtained, indicating a pre-processing loss of approximately 28.11%. Following all extraction steps, 1,725 g of native starch were recovered. This corresponds to a yield of 48.18% based on the usable fresh mass, and 34.64% relative to the total raw material.

These values are lower than those reported by Leonel and Sarmento (2008), who observed a starch yield of 53.4% using the aqueous extraction method for *A. xanthorrhiza*. The reduced efficiency observed in the present study may be attributed to several factors, including variations in tuber, post-harvest storage conditions, and differences in the mechanical and operational parameters of the extraction process, as highlighted by Daiuto et al. (2005) and more recently by Kaur et al. (2023) and Nayak et al. (2022). These authors emphasize that starch recovery is strongly influenced by the botanical source, enzyme activity, and mechanical cell disruption efficiency, all of which can affect starch liberation and purity during wet extraction.

Pasting properties

The pasting properties of native and ultrasound-modified arracacha starches (at 40%, 50%, 60%, and 70% amplitudes) were evaluated using a Rapid Visco Analyzer (RVA). Significant changes were observed in viscosity and gelatinization behavior during heating and shear (Figure 1, Table 1). Granule swelling and rupture led to paste formation, with increasing viscosity proportional to the applied amplitude.

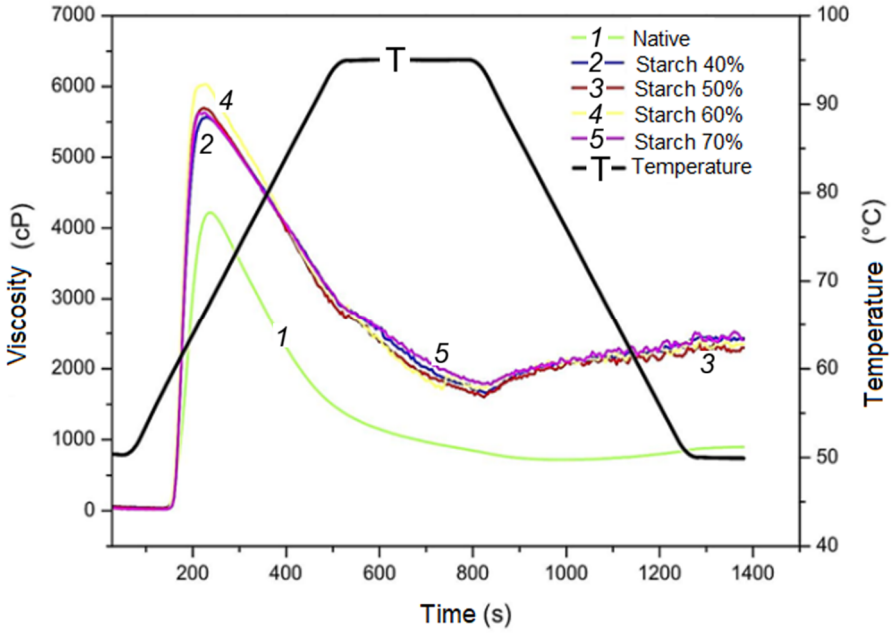

Figure 1. Viscoamylogram of native and ultrasound-treated arracacha starch

Native starch presented a pasting temperature of 78 °C and a peak viscosity (PV) of 4,000 cP. Ultrasound-treated samples showed higher pasting temperatures and PVs: 88 °C/5,500 cP (US40%), 90 °C/5,600 cP (US50%), 94 °C/6,000 cP (US60%), and 89 °C/5,550 cP (US70%). These results suggest improved thermal stability and water retention capacity, particularly at 50% and 60% amplitudes.

Table 1

Rapid Visco Analyzer (RVA) pasting properties of native and ultrasound-treated arracacha starches

Starch	Pasting temperature, °C	Peak viscosity, cP	Breakdown, cP	Retrogradation	Final viscosity, cP
Native	59.83± 0.01[a]	4284±3.51[e]	3570±1.52[e]	185±2.51[d]	904±4.00[e]
Starch 40%	58.60±0.01[b]	5568±3.51[d]	3908±1.17[c]	771±7.02[a]	2429±4.16[a]
Starch 50%	59.01±0.60[b]	5692±3.00[b]	4092±2.51[b]	698±3.00[b]	2294±3.51[d]
Starch 60%	58.61±0.01[b]	6029±4.04[a]	4322±3.00[a]	636±4.04[c]	2345±1.52[c]
Starch 70%	58.94±0.04[b]	5630±1.52[c]	3849±2.51[d]	630±4.01[c]	2414±4.50[b]

[abc]Different letters in the same column represent significant differences according to Tukey's Test ($p<0.05$)

Final viscosities (FV) were higher than their respective PVs in all modified samples, indicating enhanced molecular rearrangement during cooling. The US40% sample showed the lowest breakdown, suggesting greater resistance to thermal shear.

Reduced retrogradation tendency was observed in the ultrasound-treated samples, especially US40%, likely due to partial disruption of amorphous regions and preservation of crystalline domains (Zhang et al., 2021). Wang et al. (2022) reported similar findings in ultrasound-treated rice starches, which showed increased thermal stability and reduced setback. During RVA analysis, the sample was maintained at 90 °C for 6 minutes, followed by cooling at 5 °C/min to 50 °C, enabling setback evaluation. Gel formation and retrogradation behavior are directly influenced by amylose content, as shown by Li et al. (2023).

Thermogravimetric analysis (TGA)

The thermal degradation behavior of native and ultrasound-modified arracacha starches was evaluated by thermogravimetric analysis (TGA) and derivative thermogravimetry (DTG) (Figure 2, Table 2). Starch depolymerization typically begins above 300 °C and can be confirmed by the DTG peak temperature (Tp), corresponding to the second mass loss event (Zhang et al., 2020).

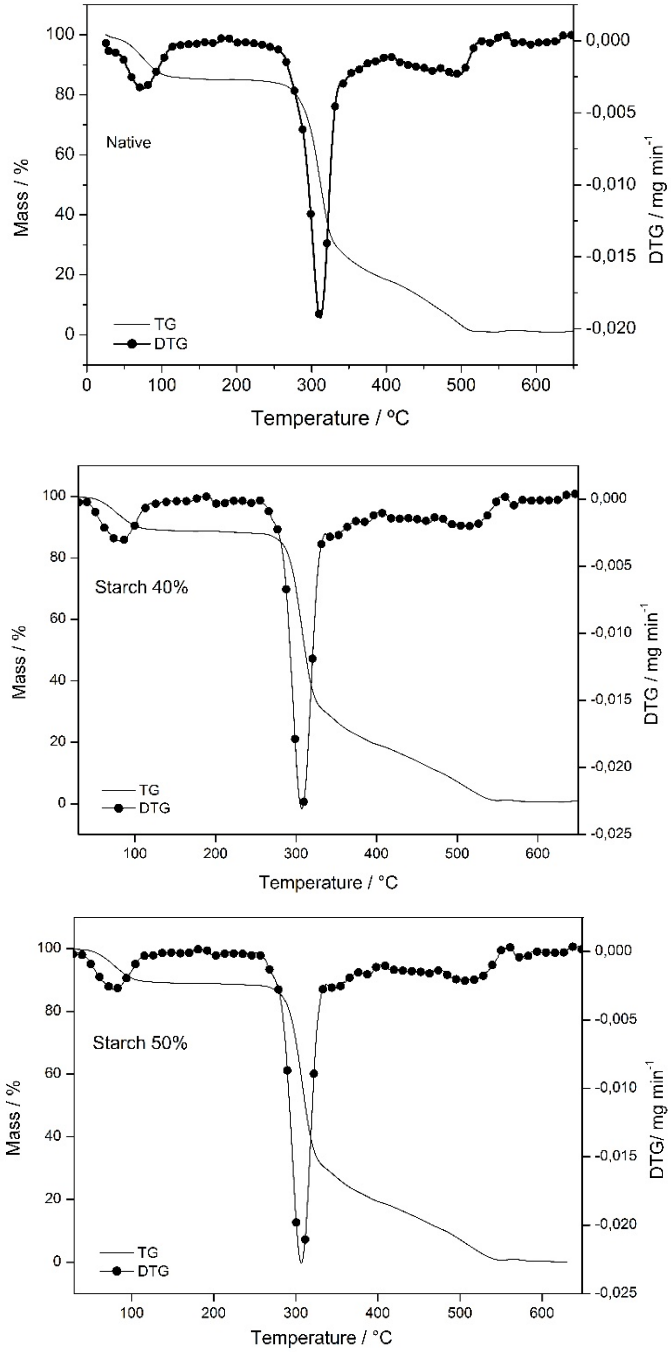

Figure 2. Thermogravimetry (TG) and Derivative Thermogravimetry (DTG) curves for the native starch, 40, 50, 60 and 70% modified starches

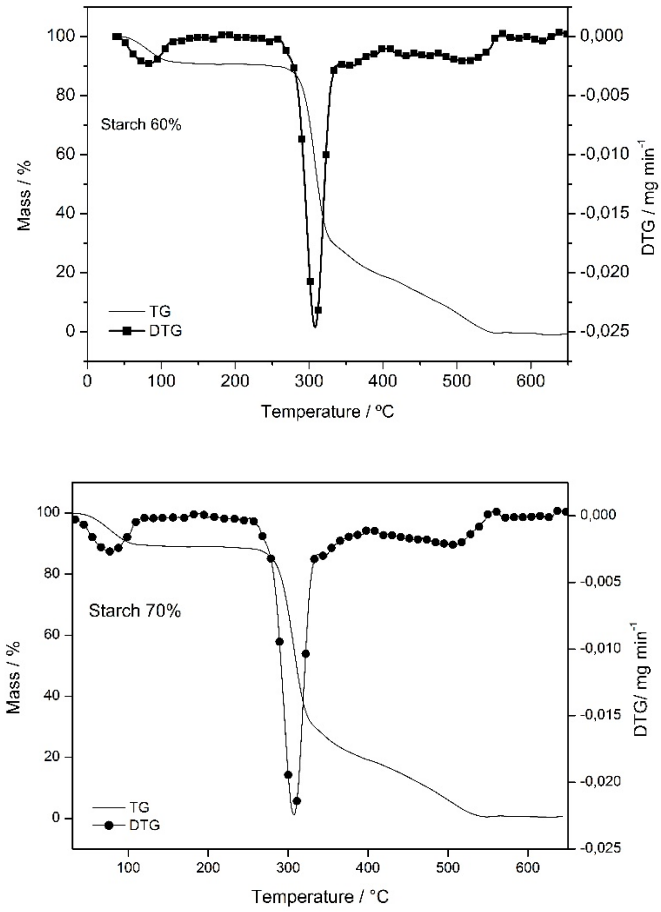

Figure 2 (*continue*). Thermogravimetry (TG) and Derivative Thermogravimetry (DTG) curves for the native starch, 40, 50, 60 and 70% modified starches

In the first mass loss event, attributed to moisture evaporation, all samples exhibited an initial temperature (Ti) of 30 °C. However, the samples treated at 40% and 50% amplitude showed the highest final temperatures (Tf), reaching 139.77 °C and 139.28 °C, respectively, indicating greater moisture retention.

In the second event, related to starch decomposition, the native starch showed the lowest Ti (248.59 °C), while the sample treated at 60% amplitude had the highest (254.76 °C), suggesting enhanced thermal resistance. In the third degradation step, the US40% sample exhibited the highest Ti (382.38 °C), while the US50% sample reached the highest Tf (596.44 °C).

The widest thermal stability range was observed for the US60% sample (137.12–254.76 °C), indicating that moderate ultrasound treatment may enhance structural reorganization. Ultrasonic cavitation likely disrupts covalent bonds in amylose and amylopectin chains and promotes new interactions, resulting in increased thermal resistance (Kumar et al., 2023).

Table 2
Thermogravimetric Analysis (TGA) and Derivative Thermogravimetry (DTG) results of native and ultrasound-modified starches

Starch	T, °C	1°	Stability	2°	3°
Native	Ti-Tf, °C Tp, °C Δm,%	30-137.19 74.78 14.10	137.19–248.59	248.59-386.10 311.88 66.15	386.10– 585.22 486.96 18.85
Starch 40%	Ti-Tf, °C Tp, °C Δm,%	30–139.77 82.90 10.60	139.77–254.24	254.24 –382.38 306.99 68.10	382.38 – 589.47 506.83 20.80
Starch 50%	Ti-Tf, °C Tp, °C Δm,%	30–139.28 77.53 10.30	139.28–253.87	253.87–356.92 307.67 63.80	356.92–596.44 562.56 25.60
Starch 60%	Ti-Tf ,°C Tp, °C Δm,%	30–137.12 84.14 9.13	137.12–254.76	254.76–380.42 308.10 69.42	380.42–571.44 507.34 21.12
Starch 70%	Ti-Tf, °C Tp, °C Δm,%	30–137.61 77.82 10.90	137.61–250.99	250.99–351.50 307.35 65.20	351.50-572.16 559.85 23.60

Δm, mass loss (%); Ti, initial temperature; Tf, final temperature; ΔT, temperature difference (°C); Tp, peak temperature (°C). Values followed by the same letter in the same column do not differ significantly according to Tukey's test ($p < 0.05$).

The greatest initial degradation (lowest Ti) and highest mass loss (Δm = 18.76%) were observed in the native starch, while ultrasound-treated samples exhibited greater thermal stability. Ash content decreased in modified samples: native (0.90%), US40% (0.50%), US50% (0.35%), US60% (0.33%), and US70% (0.30%).

Instrumental and sample-related factors, such as furnace heating rate, atmosphere, crucible geometry, particle size, and thermal conductivity, can influence TGA results (Ionashiro et al., 2012). DTG, based on the first derivative of the TGA curve over time, enhances resolution of mass loss steps, enabling precise identification of decomposition stages and thermal stability profiles in starch-based biopolymers (Aggarwal and Dollimore, 1998).

Differential Scanning Calorimetry (DSC)

Differential Scanning Calorimetry (DSC) is a thermoanalytical technique used to evaluate heat-induced physical and chemical transitions by measuring enthalpy variation (ΔH) between a sample and a reference under a programmed temperature increase (Chakraborty et al., 2022). The onset (To), peak (Tp), and conclusion (Tc) temperatures are directly related to starch granule crystallinity, X-ray diffraction patterns, and the amylose-to-amylopectin ratio, which vary significantly in modified starches (Li, 2022).

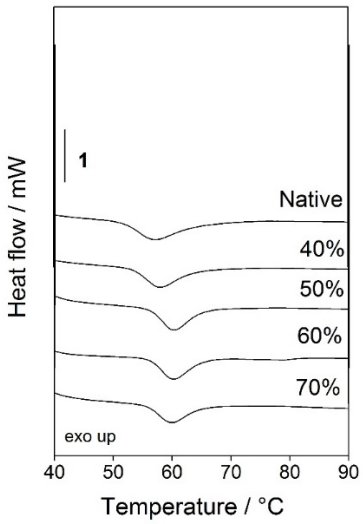

Figure 3. Differential Scanning Calorimetry (DSC) results of arracacha starch curves for the native starch, 40%, 50%, 60% and 70% modified starches

Starch gelatinization generally occurs in the presence of sufficient water and under heat (Figure 3). Table 3 reports the thermal properties during gelatinization of arracacha starch. Although the gelatinization enthalpies (ΔH_{gel}) did not differ significantly between samples (Tukey's test, $p < 0.05$), the starch treated with 40% ultrasound amplitude exhibited the lowest enthalpy (12.61 ± 0.07 J/g), indicating lower energy consumption. In contrast, the sample treated at 50% amplitude had the highest value (14.80 ± 0.19 J/g).

Wei et al. (2023a) observed similar trends in quinoa and maize starches subjected to ultrasound treatment, where prolonged exposure led to reduced gelatinization temperatures and ΔH values. These effects are attributed to cavitation-induced microjets that alter non-starch matrix components and affect starch–water interactions.

Table 3

Differential Scanning Calorimetry (DSC) results of arracacha starch

Starch	To, °C	Tp, °C	Tc, °C	ΔH, J/g
Native	59.04±0.13[a]	63.77±0.00[a]	69.14±0.38[a]	13.74±0.47[b]
Starch 40%	56.55±0.05[b]	60.96±0.01[b]	65.93±0.10[b]	12.61±0.07[c]
Starch 50%	56.12±0.02[d]	60.28±0.03[d]	65.03±0.04[c]	14.80±0.19[a]
Starch 60%	56.28±0.00[d]	60.26±0.02[d]	65.21±0.03[c]	12.82±0.11[c]
Starch 70%	55.49±0.01[c]	59.90±0.08[c]	64.73±0.01[c]	13.56±0.10[b]

[abc]Different letters in the same column represent significant differences according to Tukey's Test ($p<0.05$).

The starches evaluated in this study showed higher ΔH values than other starches, such as organic amaranth (5.92–7.17 J/g) and enzymatically treated organic rice starch (2.60–5.10 J/g), indicating denser crystalline structures and greater thermal stability.

In studies involving hydrocolloids, gelatinization onset temperature (To) either increases or remains stable, depending on molecular interactions (Chakraborty et al., 2022). For instance, interactions between corn starch and gums such as xanthan, guar, and carboxymethylcellulose can increase due to water competition and steric hindrance. Gelatinization temperature ranges for arracacha starches were narrower (7.00–7.39 °C) than those of common starches like potato (~20 °C), suggesting more uniform and stable crystalline domains (Li, 2022). This narrow range reflects improved crystal perfection and homogeneity, reinforcing the potential of ultrasound as a structural modification tool.

Field Emission Gun – Scanning Electron Microscopy (FEG-SEM)

Field Emission Gun - Scanning Electron Microscopy (FEG-SEM) was used to evaluate the morphology of native and ultrasound-treated arracacha starch granules. All samples exhibited polygonal shapes with visible concentric growth rings, typical of structured granules (Figure 4).

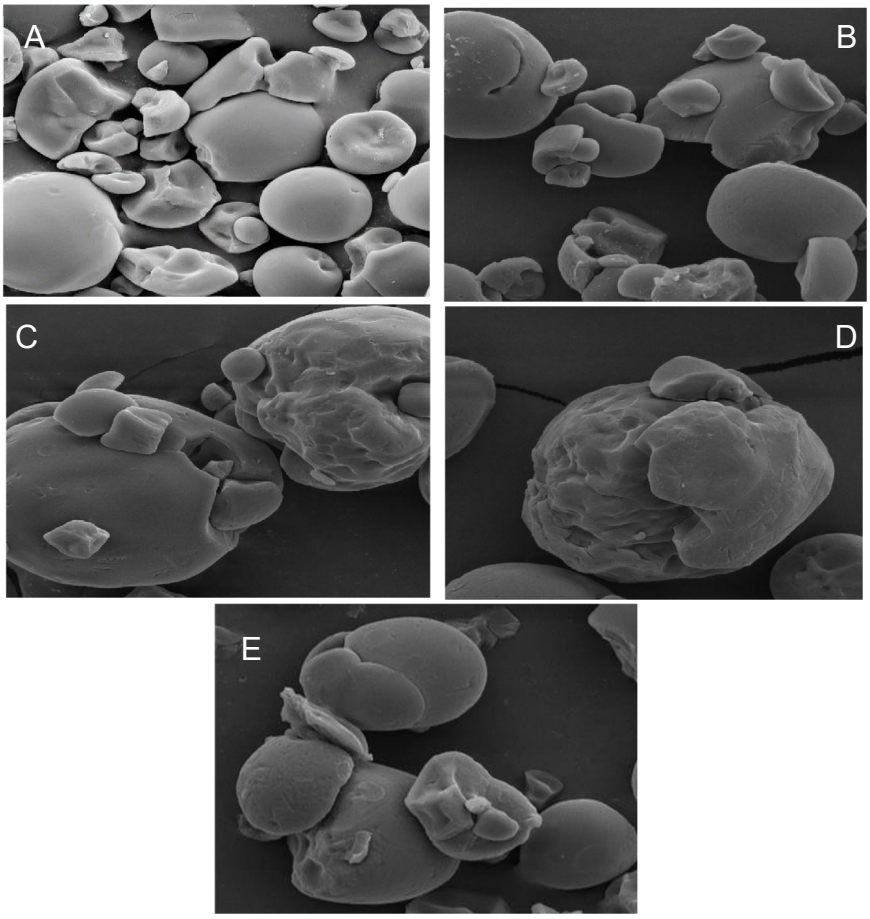

Figure 4. Scanning electron micrographs (5,000×) of native starch and starches modified by ultrasound at 40%, 50%, 60%, and 70% amplitudes:
A – Native starch, B – Starch modified at 40% amplitude;
C – Starch modified at 50% amplitude; D – Starch modified at 60% amplitude;
E – Starch modified at 70% amplitude.

The average granule size increased from 12.27 µm (native) to 17.77 µm at 50% ultrasound amplitude, followed by slight reductions at 60% and 70% amplitudes, suggesting surface erosion at higher intensities.

No granule agglomeration was observed, indicating that the ultrasound conditions applied were effective and did not compromise granule dispersion. These results are

consistent with recent findings in pea and maize starches, where ultrasound treatment caused surface roughness, microcracks, and limited swelling without loss of structural integrity (Vela et al., 2024; Wei et al., 2023b).

Ultrasound primarily disrupts amorphous regions of the starch granule, preserving crystallinity while enhancing porosity—properties that can be advantageous for applications requiring increased water interaction or enzymatic access. The observed morphological changes support the potential of ultrasonic treatment as a controlled physical modification method for starch-based systems (Jamlabadi et al., 2019; Leonel et al., 2008).

X-ray diffraction (XRD)

X-ray diffraction (XRD) analysis reveals that starch granules exhibit a lamellar structure with nanometric subunits (~9–10 nm), primarily attributed to amylopectin. This crystalline region is densely packed, hindering water and enzyme penetration and increasing resistance to hydrolysis (Zhang et al., 2023). The crystallinity patterns are classified into types A, B, and C, corresponding to the double helix packing of branched chains in amylopectin: type A is typical of cereals such as maize and rice; type B predominates in tubers and retrograded starch; and type C is an intermediate form common in roots and legumes.

The relative crystallinity index (RCI), quantified by XRD, is crucial to understanding starch structural stability (Table 4).

Table 4

Crystallinity and Diffraction Pattern Peaks

Starch	Relative crystallinity,%	Standard deviation
Native	26.34	0.0100
Starch 40%	24.03	0.0100
Starch 50%	24.71	0.0110
Starch 60%	25.67	0.0039
Starch 70%	25.86	0.0032

Crystallinity directly influences water interactions and resistance to chemical and enzymatic treatments, although excessive crystallinity may cause brittleness (Wang et al., 2021). In this study, ultrasound treatment caused changes in crystallinity depending on amplitude, with reductions at low intensity (40%) and increases at moderate amplitudes, indicating structural reorganization and a biphasic effect of ultrasound (Yu et al., 2024; Zhang et al., 2023).

Freeze-thaw cycles significantly affect starch crystalline structure, increasing amorphous regions while reducing crystalline fractions, which affects functional properties (Liu et al., 2020; Wang et al., 2019). Ultrasound primarily acts on the amorphous regions, causing localized granule disruption without altering starch polymorphs, which is beneficial for functional modification while maintaining the fundamental structure (Zheng et al., 2013).

Therefore, controlled ultrasound application emerges as a promising technique to modulate starch crystallinity and physicochemical properties, optimizing its industrial applications that require specific traits such as enhanced mechanical resistance or water absorption capacity (Vela et al., 2024).

Conclusions

The aqueous extraction of mandioquinha-salsa starch yielded results consistent with previous studies, using a simple, safe, and industrially applicable method. Ultrasonic treatment at 20 kHz significantly modified the starch properties, as confirmed by RVA, TG/DTG, DSC, FEG-SEM, and X-ray diffraction analyses. The 40% amplitude treatment showed improved shear resistance and reduced retrogradation, whereas higher amplitudes (50% and 60%) enhanced thermal stability. Morphological changes occurred without granule agglomeration, and crystallinity increased moderately but remained below that of native starch.

Overall, ultrasound proved to be an effective, sustainable, and energy-efficient method for modifying mandioquinha-salsa starch, broadening its potential in the food industry. Its low gelatinization temperature and high peak viscosity make it particularly suitable for products requiring gelation and refrigeration. This study also contributes new insights into the ultrasonic modification of this starch source, highlighting ultrasound as a clean and viable alternative for starch processing.

Acknowledgment. The authors would like to thank the Brazilian agencies CAPES and CNPq for their financial support, and the State University of Ponta Grossa (UEPG) for institutional support.

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Cite:

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