AGROCIENCIA URUGUAY

Agrociencia Uruguay 2026 | Volume 30 | Number NE1 | Article e1626
DOI: 10.31285/AGRO.30.1626
ISSN 2730-5066

Investigaciones en pecán en Sudamérica

Optimizing Particle Size to Enhance Phenolic Compound Recovery From Pecan Shells for Bioactive Applications

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Abstract

The global production of pecan nuts (*Carya illinoinensis*) is expanding, generating growing interest in the valorization of their by-products, particularly the shell, a promising source of bioactive compounds and natural antioxidants. To support its use in food, nutraceutical, and pharmaceutical applications, optimizing extraction conditions is essential. In this study, pecan shells were dried, ground, and sieved into three particle sizes: coarse (250-150 µm), medium (150-100 µm), and fine (≤100 µm). Phenolic compounds were extracted using an ultrasonic bath, and total phenolics were quantified by the Folin-Ciocalteu method. Reducing power and the ability of the extracts to scavenge DPPH and ABTS radicals were also assessed. LC-MS/MS was used to characterize the most efficient extract. Fine particles produced higher extract yields, while medium and fine particles resulted in greater total phenolic content. Twelve phenolic compounds were identified, including the flavonoids gallocatechin 3-O-gallate, catechin, gallocatechin, procyanidin C1, procyanidin B1, isorhamnetin 3-glucuronide, and taxifolin 3-O-rhamnoside, as well as methyl chlorogenate, gallic acid 3-O-gallate, and 3-caffeoylquinic acid. Antioxidant activity increased with particle size reduction down to 100 µm, but smaller sizes reduced this activity. Extracts obtained from medium particles showed the highest phenolic content (20.52 ± 0.08 mg GAE/g) and antioxidant activity (58.81 ± 0.80% for DPPH and 79.00 ± 0.96% for ABTS). These findings provide valuable insights into optimizing phenolic extraction from pecan shells for applications in food and health-related industries.

Keywords: mass spectrometry, radical scavenging, phytochemicals, natural antioxidants, liquid extracts

Editor

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Received *9 Dec 2024*
Accepted *22 May 2025*
Published *13 Feb 2026*

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Siebeneichler, T. J., Peixoto, E. C., Hoffmann, J. F., Rombaldi, C. V., & Dias, A. R. G. (2026). Optimizing particle size to enhance phenolic compound recovery from pecan shells for bioactive applications. *Agrociencia Uruguay, 30*(NE1), Article e1626. https://doi.org/10.31285/AGRO.30.1626

Optimización del tamaño de partícula para la extracción de compuestos fenólicos de cáscaras de pecán

Resumen

La producción global de nueces pecán (*Carya illinoinensis*) está en expansión, lo que ha generado un creciente interés en la valorización de sus subproductos, especialmente la cáscara, reconocida como una fuente prometedora de compuestos bioactivos y antioxidantes naturales. Para apoyar su uso en aplicaciones alimentarias, nutracéuticas y farmacéuticas, es esencial optimizar las condiciones de extracción. En este estudio, las cáscaras fueron secadas, molidas y tamizadas en tres tamaños de partícula: gruesa (250-150 µm), mediana (150-100 µm) y fina (≤100 µm). Los compuestos fenólicos fueron extraídos mediante baño ultrasónico y cuantificados por el método de Folin-Ciocalteu. También se evaluaron el poder reductor y la capacidad de los extractos para eliminar radicales DPPH y ABTS. La técnica LC-MS/MS se utilizó para caracterizar el extracto más eficiente. Las partículas finas produjeron mayores rendimientos de extracto, mientras que las partículas medianas y finas generaron un mayor contenido fenólico total. Se identificaron doce compuestos fenólicos, incluidos los flavonoides galocatequina 3-O-galato, catequina, galocatequina, procianidina C1, procianidina B1, isoramnetina 3-glucurónido y taxifolina 3-O-ramnósido, así como metil clorogenato, ácido gálico 3-O-galato y ácido 3-cafeoilquínico. La actividad antioxidante aumentó con la reducción del tamaño de partícula hasta 100 µm, pero tamaños menores redujeron esta actividad. Los extractos de partículas medianas mostraron el mayor contenido fenólico (20,52 ± 0,08 mg GAE/g) y actividad antioxidante (58,81 ± 0,80 % para DPPH y 79,00 ± 0,96 % para ABTS). Estos resultados aportan información valiosa para optimizar la extracción de compuestos fenólicos de cáscaras de nuez pecán para aplicaciones en la industria alimentaria y de la salud.

Palabras clave: espectrometría de masas, captura de radicales, fitoquímicos, antioxidantes naturales, extractos líquidos

Otimização do tamanho de partícula para a extração de compostos fenólicos de cascas de pecã

Resumo

A produção global de nozes-pecã (*Carya illinoinensis*) está em expansão, gerando crescente interesse na valorização de seus subprodutos, especialmente a casca, reconhecida como uma fonte promissora de compostos bioativos e antioxidantes naturais. Para ampliar seu uso em aplicações alimentícias, nutracêuticas e farmacêuticas, é essencial otimizar as condições de extração. Neste estudo, as cascas foram secas, moídas e peneiradas em três tamanhos de partícula: grosso (250-150 µm), médio (150-100 µm) e fino (≤100 µm). Os compostos fenólicos foram extraídos em banho ultrassônico e quantificados pelo método de Folin-Ciocalteu. Avaliaram-se também o poder redutor e a capacidade dos extratos de sequestrar radicais DPPH e ABTS. A técnica LC-MS/MS foi empregada para caracterizar o extrato mais eficiente. As partículas finas produziram maior rendimento de extrato, enquanto partículas médias e finas apresentaram maior teor fenólico total. Doze compostos fenólicos foram identificados, incluindo os flavonoides galocatequina 3-O-galato, catequina, galocatequina, procianidina C1, procianidina B1, isoramnetina 3-glucuronídeo e taxifolina 3-O-ramnosídeo, além de metil clorogenato, ácido gálico 3-O-galato e ácido 3-cafeoilquínico. A atividade antioxidante aumentou com a redução do tamanho de partícula até 100 µm, mas tamanhos menores reduziram essa atividade. Extratos de partículas médias apresentaram o maior teor fenólico (20,52 ± 0,08 mg GAE/g) e a maior atividade antioxidante (58,81 ± 0,80% para DPPH e 79,00 ± 0,96% para ABTS). Esses resultados fornecem informações relevantes para otimizar a extração de compostos fenólicos de cascas de noz-pecã nas indústrias alimentícia, nutracêutica e farmacêutica.

Palavras-chave: espectrometria de massas, sequestro de radicais, fitoquímicos, antioxidantes naturais, extratos líquidos

1. Introduction

The pecan (*Carya illinoinensis*) is a widely cultivated nut tree in North America, known for its edible kernels (Siebeneichler et al., 2023). However, the shells, which constitute a substantial portion of the nut's mass, are often discarded as waste. Pecan shells are a byproduct of nut processing and are composed of hemicellulose, cellulose, and lignin. This structural composition makes the shell hard, non-toxic, biodegradable, and renewable. Between 0.460 and 0.486 kg of shell is generated per kilogram of pecans (Hoffmann et al., 2024). Typically, these shells are either discarded into the environment or burned, but they could be utilized more efficiently.

Several studies have identified pecan shells as a rich source of phenolic compounds, such as phenolic acids, flavonoids, and tannins (Hilbig et al., 2018; Kureck et al., 2018; Reyes-Vázquez et al., 2022; Villasante et al. 2019), which exhibit strong antioxidant activity (Flores-Estrada et al., 2020; Villasante et al. 2019; Villasante Dueñas, 2021), antimicrobial (Caxambú et al., 2016; Neira-Vielma et al., 2022; Villasante et al., 2020), and antifungal properties (Caxambú et al., 2016; García-García et al., 2025). These bioactivities make them promising candidates for natural food preservation and health-promoting formulations. Antioxidants play a crucial role in preventing oxidative stress, which is implicated in the pathogenesis of various chronic diseases, including cancer, cardiovascular diseases, and neurodegenerative disorders (Ottaviani et al., 2018; Sunil & Xu, 2019). As the demand for natural antioxidants in the food industry grows, agricultural and food waste is increasingly becoming an ideal source for the extraction of phenolic compounds as natural antioxidants. Currently, there is a growing research effort focused on recycling low-cost waste products from the food, forestry, and agricultural industries, due to both environmental and economic benefits (Pontonio et al., 2024).

The performance of phenolic compound extraction from plant materials is influenced by various external parameters, such as temperature, time, and solid-to-solvent ratio. Among these, particle size reduction is a key pre-treatment that increases the surface area available for solvent interaction, potentially enhancing both extract yield and the recovery of bioactive compounds (Hilbig et al., 2018; Yusoff et al., 2022). However, studies on the impact of particle size on phenolic extraction from pecan shells remain limited (Prasedya et al., 2021). Previous studies with shells have evaluated different solvents, temperatures, and extraction methods, with ultrasound-assisted extraction under heating showing superior results compared to conventional extractions (Hilbig et al., 2018). However, the concentration and extraction of phenolic compounds relative to exposed surface area are not well documented. Evidence from other plant matrices, such as barley (Zhou et al., 2021), plant leaves (Alsaud & Farid, 2020), and apple (Kim et al., 2017), shows that while finer particles can improve extraction, overly fine milling may also cause compound degradation due to enzymatic oxidation or excessive interactions with matrix components (Alsaud & Farid, 2020; Kim et al., 2017).

Reducing particle size can increase the surface area available for solvent penetration, potentially improving the efficiency of polyphenol extraction and the overall yield of the extract (Prasedya et al., 2021). Previous studies have evaluated different solvents, temperatures, and extraction methods, with ultrasound-assisted extraction under heating showing superior results compared to conventional extractions (Hilbig et al., 2018). However, the concentration and extraction of phenolic compounds relative to exposed surface area are not well documented.

In this context, this study aims to evaluate the effect of particle size reduction on ultrasound-assisted extraction, as well as the yield and antioxidant activity of phenolic compounds from pecan shells. Establishing such a relationship between pecan shell particle size, phenolic extractability, and antioxidant properties will provide valuable insights for the development of more effective and sustainable methods for utilizing this byproduct.

2. Materials and Methods

2.1 Chemicals

Ethanol P.A., methanol P.A., and Folin-Ciocalteu reagent were obtained from Êxodo Científica, and gallic acid was sourced from Dinâmica Química. DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2-azino-bis(3-ethylbenzothiazoline)-6-sulfonic acid) were purchased from Sigma-Aldrich.

2.2 Materials

'Barton' pecan nutshells were provided by Divinut, located in Cachoeira do Sul, RS, Brazil, in April 2024.

2.3 Sample Preparation

The shells were selected and dried in a forced-air (Ethik, Canadá) oven for 24 hours at 45 °C. The shells were cleaned, dried, and milled into various particle sizes using a mechanical grinder (Garthen, Brazil). Subsequently, the milled material was classified into three particle size groups using an electromagnetic sieve shaker (Bertel, Brazil). The first sample was collected between the 60 and 100 mesh sieves, corresponding to the coarse particle size (250 µm-150 µm). The second sample was obtained between the 100 and 150 mesh sieves, representing the medium particle size (150 µm-100 µm). Finally, the third sample was collected from the 150 mesh sieve, characterizing the fine particle size (≤100 µm) (Figure 1).

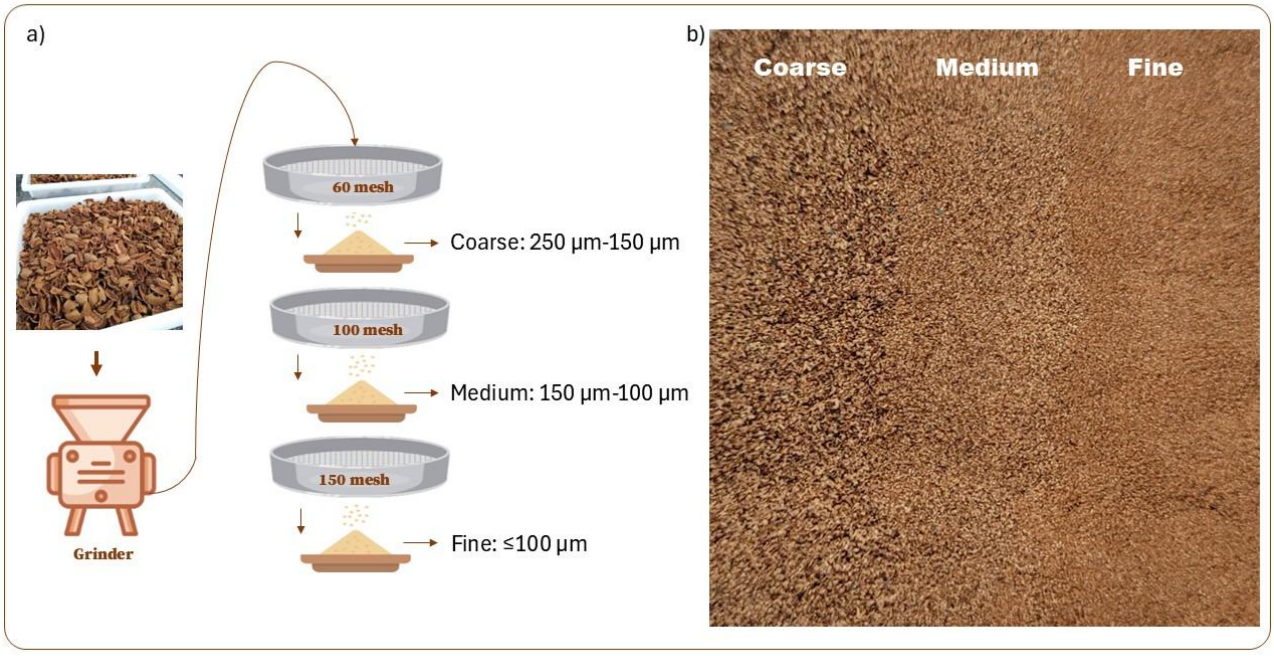


Figure 1. (a) First sample collected between the 60 and 100 mesh sieves, corresponding to the coarse particle size (250 µm-150 µm). Second sample obtained between the 100 and 150 mesh sieves, representing the medium particle size (150 µm-100 µm). Finally, third sample collected from the 150 mesh sieve, characterizing the fine particle size (≤100 µm). Each fraction was analyzed. (b) Photographs of ground pecan shell treatments

2.4 Extraction of Polyphenols

For the extraction of polyphenols from pecan nutshells, dry and ground samples were utilized, subjected to a solid-liquid extraction method based on the methodology of Hilbig et al. (2018), with adaptations. Initially, 1.0 g of pecan shell powder was placed in a conical flask, to which 30 mL of 20% ethanol was added to facilitate the extraction of the compounds. The mixture was vortexed for 30 seconds to ensure adequate homogenization.

Subsequently, extraction was conducted in an ultrasonic bath (Cristófoli, Brazil) at 60 °C, operating at 52 kHz and a frequency of 60 Hz, for 35 minutes. After sonication, the resulting extract was centrifuged at 4,000 rpm for 10 minutes (Eppendorf 5430 R centrifuge, Germany) to separate the solid fractions. The obtained supernatant was then stored in a freezer at −20 °C.

2.5 Determination of Extract Yield

The yield of the polyphenol extract was calculated as a percentage of the initial dry weight of the pecan shell powder. The dried extract was weighed after solvent evaporation, and the yield was expressed as:

$$Yield\ (\%) = \frac{\text{Weight of the dried extract (g)}}{\text{Weight of the initial dry sample (g)}} \times 100$$

2.6 Determination of Total Phenolic Content (TPC)

The TPC was determined using the Folin-Ciocalteu method by Swain and Hillis (1959). Initially, the extracts were diluted by pipetting a 40 µL aliquot of extract into 1960 µL of distilled water. After dilution, 0.250 mL of the diluted extract and 4 mL of distilled water were mixed with 0.250 mL of Folin-Ciocalteu reagent, vortexed for 10 seconds, followed by a 3-minute reaction, and then 0.5 mL of a 1 mol.L^{-1} sodium carbonate solution was added. The mixture was vortexed for another 10 seconds. After 1 hour of reaction in the dark, absorbance was measured using a UV-Vis spectrophotometer (Tecnal UV-5100 model, Metash, Shanghai, China). Quantification was performed using a calibration curve prepared with gallic acid, at concentrations ranging from 5 to 150 µg.mL^{-1} ($y = 0.0042x - 0.016$, $R^2 = 0.99$). The total phenolic content was expressed as milligrams of gallic acid equivalents (mg GAE/g).

2.7 Antioxidant Activity Assay (DPPH)

The antioxidant activity of the extracts was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay. A 0.1 mM DPPH solution in methanol was prepared, and 3.9 mL of this solution was added to 100 µL of the extract (previously diluted 60-fold) at various concentrations. The mixture was shaken vigorously and incubated in the dark for 2 hours. Absorbance was measured at 515 nm, and the percentage of DPPH radical inhibition was calculated.

$$DPPH\ inhibition\ (\%) = \frac{\text{A control} - \text{A Sample}}{\text{A Control}} \times 100$$

2.8 Antioxidant Activity Assay - ABTS Radical Scavenging Assay

The ABTS radical scavenging assay was performed following the method described by Re et al. (1999). The ABTS radical cation (ABTS$^{\cdot+}$) was generated by mixing 7.0 mM ABTS solution with 2.45 mM potassium persulfate solution in a 1:1 (v/v) ratio. The mixture was kept in the dark at room temperature for 16 hours to complete the reaction. The resulting solution was then diluted with ethanol to achieve an absorbance of 0.70. For the assay, 30 µL of phenolic extract (previously diluted 60-fold) was mixed with 3 mL of the ABTS working solution. An equal amount of ethanol (30 µL) was used as a blank (control). All samples were prepared in triplicate and vortexed for 30 seconds. After 6 minutes of incubation, the absorbance decrease was measured against ethanol as a blank using a UV-Vis spectrophotometer (Tecnal UV-5100 model, Metash, Shanghai, China) at 734 nm. The percentage of ABTS inhibition was calculated using the formula:

$$ABTS\ inhibition = \frac{\text{A control} - \text{A Sample}}{\text{A Control}} \times 100$$

2.9 Determination of Individual Phenolic Compounds

LC-MS/MS analyses were conducted using a liquid chromatography system coupled with a QToF mass spectrometer (Shimadzu, model LCMS-9030). Metabolite separation was performed on a C18 column with water containing 0.1% formic acid (solvent A) and acetonitrile with 0.1% formic acid (solvent B) in a gradient program started at 5% solvent B, increased linearly to 90% over 15 minutes, maintained for 3 minutes, then returned to 5% over 2 minutes, and held for an additional 6 minutes. Mass spectrometry conditions were set in negative ionization mode, covering a mass range of 150 to 950 m/z. Calibration was performed with 2.6 mM sodium iodide, and data were processed using LabSolutions Insight software.

2.10 Statistical Analysis

All experiments were conducted in triplicate, and results were expressed as mean ± standard deviation. Statistical analysis was performed using ANOVA followed by Tukey's test to determine significant differences between groups. A p-value of <0.05 was considered statistically significant.

3. Results

The results of pecan shell grinding into different particle sizes are presented in Figure 2 and Table 1. The analyses of antioxidant activity (DPPH and ABTS), total phenolic content (TPC), and extract yield revealed significant variations among the different treatments.

3.1 Extract Yield

The fine particles showed the highest yield (25.90 ± 2.96%). However, there was no significant difference between the yield of the medium particles (14.64 ± 1.46%) and the coarse particles (11.78 ± 2.33%) (Figure 2).

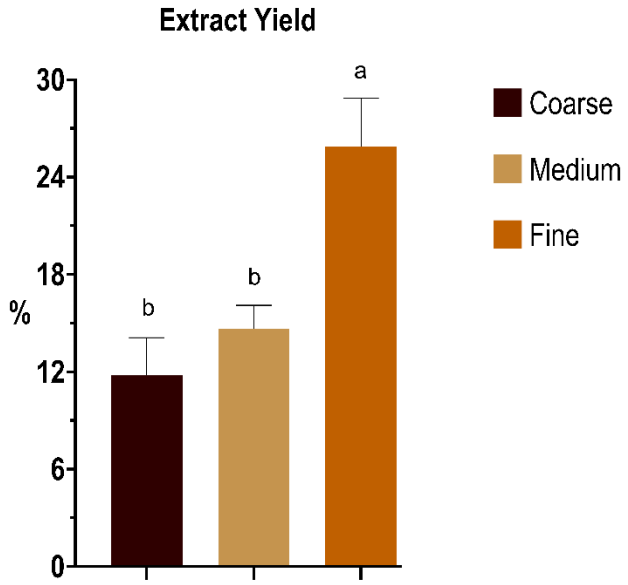


Figure 2. Yield of extracts from pecan nutshells at varying particle sizes

3.2 Total Phenolic Content

The phytochemical composition of pecan nutshell extract exhibited a higher TPC of 20.52 ± 0.08 mg GAE/g in samples with medium particle sizes, followed by fine particles at 18.31 ± 0.67 mg GAE/g, while coarse particles demonstrated the lowest TPC at 12.65 ± 1.86 mg GAE/g (Table 1).

3.3 Antioxidant Activity

The results of the antioxidant activity, assessed through DPPH and ABTS assays, showed significant variations based on the different particle sizes of the pecan nutshell. The extracts from medium-sized particles displayed the best results, with an inhibition of 58.81 ± 0.80% in the DPPH assay and 79.00 ± 0.96% in the ABTS assay. In contrast, the fine particle extracts exhibited an inhibition of 47.33% in DPPH and 70.22% in ABTS, surpassing only the coarse particle extracts in both tests. These results indicate that medium particle size milling is more effective for maximizing antioxidant activity.

3.4 Profile of Phenolic Compounds in Pecan Nutshell Extracts

The extracts with medium particle size (150 µm-100 µm) were analyzed using HPLC-ESI-QTOF-MS to identify the phenolic compounds extracted from the pecan shell. A total of 12 phenolic compounds were identified, specifically within the flavonoid group, including gallocatechin 3-O-gallate, catechin, gallocatechin, procyanidin C1, procyanidin B1, isorhamnetin 3-glucuronide, and taxifolin 3-O-rhamnoside. In the class of phenolic acids, methyl chlorogenate, gallic acid 3-O-gallate, and 3-caffeoylquinic acid were found. Additionally, the compound 3,7-Dihydroxy-8-methoxy-5,10-dioxo-5,10-dihydrochromeno[5,4,3-cde]chromen-2-yl beta-D-xylopyranoside and phloridzin were also identified (Table 2).

Table 1. Antioxidant activity and total phenolic content of different particle sizes

Particle size	DPPH (%)	ABTS (%)	TPC
Coarse	38.51 ± 0.91c	64.48 ± 0.40c	12.65 ± 1.86b
Medium	58.81 ± 0.80a	79.00 ± 0.96a	20.52 ± 0.08a
Fine	47.33 ± 1.55b	70.22 ± 0.77b	18.31 ± 0.67a

Table 2. Profile of phenolic compounds in pecan nutshell extracts

Compounds	RT (min)	Score	m/z (Theoretical)	[M-H]- experimental	Molecular Formula (M)
Gallocatechin 3-*O*-gallate	1.565	93.92	458.08491	457.07763	$C_{22}H_{18}O_{11}$
Catechin	6.587	94.69	290.07904	289.07176	$C_{15}H_{14}O_6$
Gallocatechin	3.264	90.62	306.07395	305.06668	$C_{15}H_{14}O_7$
Procyanidin C1	6.353	68.28	866.20581	865.19854	$C_{45}H_{38}O_{18}$
Methyl chlorogenate	1.431	60	368.11072	367.10453	$C_{17}H_{20}O_9$
Gallic acid 3-*O*-gallate	1.737		322.03248	321.02521	$C_{14}H_{10}O_9$
3-Caffeoylquinic acid	7.446	49.6	354.09508	353.08781	$C_{16}H_{18}O_9$
Procyanidin B1	6.3	97.06	578.14243	577.13515	$C_{30}H_{26}O_{12}$
3,7-Dihydroxy-8-methoxy-5,10-dioxo-5,10-dihydrochromeno[5,4,3-cde]chromen-2-yl beta-D-xylopyranoside	8.408	88.4	448.06418	447.0569	$C_{20}H_{16}O_{12}$
Isorhamnetin 3-glucuronide	8.124	39.95	492.09039	491.08311	$C_{22}H_{20}O_{13}$
Phloridzin	8.836	66.77	436.13695	435.12967	$C_{21}H_{24}O_{10}$
Taxifolin 3-*O*-rhamnoside	7.289	86.42	450.11621	449.10894	$C_{21}H_{22}O_{11}$

4. Discussion

The extraction of polyphenols from pecan nutshells is particularly relevant given the significant growth of the pecan industry in recent years across several countries, such as China (Jia et al., 2019), Australia, Israel (Ferrara et al., 2023), Brazil (Ribeiro et al., 2020), Argentina, Peru (Cambareri et al., 2023), South Africa (Achilonu et al., 2023), and Uruguay (Ferrari et al., 2022). In the industrial process, an average of 460-486 kg of shells is generated per ton of pecan nuts processed (Hoffmann et al., 2024), highlighting the abundance and importance of this by-product. Pecan nutshells are rich in phenolic compounds with known antioxidant (Moccia et al., 2020; Villasante et al., 2020; Villasante Dueñas, 2021), antifungal (Caxambú et al., 2016; García-García et al., 2025), and antimicrobial properties (Caxambú et al., 2016; Neira-Vielma et al., 2022; Villasante et al., 2020). These bioactivities make them promising natural alternatives for food preservation and functional enhancements. The recovery of such compounds aligns with the increasing demand for sustainable solutions and the reduction of synthetic additives in the food industry, offering both economic and environmental advantages.

The influence of particle size on the extraction of phenolic compounds from pecan nutshells was evaluated through three parameters: TPC, antioxidant activity (DPPH and ABTS assays), and extract yield. Our results indicated that decreasing particle size increased extraction yield, as expected, since smaller particles enhance the surface area of contact between the material and the solvent, facilitating extraction (Prasedya et al., 2021). Moreover, milling, as a physical modification, disrupts the rigid structure of the pecan shell, altering its physicochemical properties and facilitating the extraction of various compounds, not just the bioactive ones (Huang et al., 2020). In this regard, it is important to emphasize that an increase in yield does not always reflect an increase in the purity of the extract or in the concentration of bioactive compounds.

In general, previous studies indicate that the extraction of bioactive compounds is favored by reducing particle size, as demonstrated in studies involving barley (Zhou et al., 2021) and plant leaves (Alsaud & Farid, 2020). However, our results show that this effect occurs only up to a certain point. Particles with a size equal to or less than 100 µm (fine) exhibited phenolic compound content statistically equivalent to that of medium-sized particles (150 to 100 µm). Additionally, antioxidant activity was higher in medium-sized particles (Table 1). Previous studies corroborate our findings, showing that reducing particle size does not always lead to increased recovery of bioactive compounds. For example, in teas, the contents of polyphenols and catechins decreased with the reduction of particle size (Hu et al., 2012). Furthermore, in some cases, particle size characteristics do not influence the amount of bioactive compounds extracted (Cheng et al., 2021).

Moreover, the fact that fine milling did not demonstrate the highest phenolic compound content may be attributed to increased interactions between the phenolic compounds and other substances in the sample, such as proteins, lipids, and carbohydrates present in the shell. The breakdown of the cell wall may also release endogenous enzymes, such as cytoplasmic polyphenol oxidase, resulting in the oxidative degradation of phenolic compounds (Quan et al., 2020). Superfine milling of apples has shown similar results. In this case, milling at atmospheric pressure resulted in considerable darkening of the particles and a reduction in antioxidant activity, attributed to greater interaction between polyphenol oxidase and phenolic compounds due to increased oxygen access (Kim et al., 2017).

Phenolic compounds are the primary contributors to antioxidant activity in pecans (Cardona Jimenez et al., 2024; Siebeneichler et al., 2023). In this study, the ABTS assay revealed a higher percentage of inhibition compared to DPPH, even when using extracts at the same concentrations. This difference may be attributed to the greater sensitivity of the ABTS assay in detecting antioxidant activity, given its faster reaction kinetics and more intense response to antioxidants compared to other free radicals (Prasedya et al., 2021).

Based on our results, we suggest that milling to a medium particle size (150 to 100 µm) is sufficient to optimize the extraction of phenolic compounds. It is essential to consider the balance between extraction efficiency and energy costs when planning extraction processes on an industrial scale. Milling to finer particle sizes, besides not resulting in higher recovery of bioactive compounds, requires more energy and may present operational challenges, such as screen clogging, making the process less efficient in terms of cost-benefit.

The profile of phenolic compounds in the medium particle size extract was characterized by HPLC-ESI-QTOF-MS, identifying 12 phenolic compounds. Among them, flavonoids such as gallocatechin 3-O-gallate, catechin, and gallocatechin stand out, being widely recognized for their biological activities. These compounds, present in high concentrations (30%) in green tea, have been associated with various health benefits, including anti-cancer, antidiabetic, and anti-inflammatory properties (Rashidinejad et al., 2021). Additionally, procyanidin C1 and procyanidin B1 were identified, compounds that contribute to intestinal (Wei et al., 2022) and cardiovascular health (Ottaviani et al., 2018); isorhamnetin 3-glucuronide, which acts as an insulin regulator (Matboli et al., 2021); and taxifolin 3-O-rhamnoside, known for its prominent anticancer activity (Sunil & Xu, 2019). Phenolic acids such as methyl chlorogenic acid, gallic acid 3-O-gallate, and 3-caffeoylquinic acid were also identified, all associated with important biological activities (Xiang et al., 2024; Xue et al., 2023). These results are partially consistent with the literature (Hilbig et al., 2018; Kureck et al., 2018; Xu et al., 2020), with observed differences possibly attributed to cultivar variability (Frezza et al., 2025), extraction methods (Hilbig et al., 2018) and the stability of the extracted compounds (Jia et al., 2023). Our findings reinforce the bioactive potential of pecan nutshell extracts.

This study represents an important step in optimizing the extraction process of phenolic compounds from pecan nutshells. It provides a valuable foundation for researchers and industries in the food, nutraceutical, and pharmaceutical sectors, serving as a starting point in the pursuit of more efficient methods for extracting these bioactive compounds.

5. Conclusions

The particle sizes affect extract yield, phenolic compounds content and antioxidant activities. The results indicated that reducing particle size to ≤ 100 µm (fine) resulted in the highest extraction yield. However, particle with medium range (150 to 100 µm) associates better phenolic compounds content and antioxidant activity. To optimize the production process and reduce energy consumption, there is no need to reduce particle size below 100 µm.

Acknowledgements

We thank Divinut for providing the pecan nutshells, CNPq, FAPERGS (24/2551-0001497-7), (23/2551-0001828-4), and support by PPGCTA/UFPel.

Transparency of data

Available data: The entire data set that supports the results of this study was published in the article itself.

Author contribution statement

	TF Siebeneichler	EC Peixoto	JF Hoffmann	CV Rombaldi	ARG Dias
Conceptualization	■				■
Funding acquisition					■
Investigation	■	■			
Writing – original draft	■	■			
Writing – review and editing			■	■	■

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