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Valorization of *Garcinia humilis* Residues in Active Collagen/Starch Films for Biodegradable Packaging Applications

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ABSTRACT

This study developed and characterized biodegradable films of collagen and starch of sweet potato incorporated with phenolic extract of *Garcinia humilis* residue extract (GHRE), aiming at their application as active packaging. GHRE showed a high content of total phenolics (543.69 mg GAE/g), with outstanding antioxidant activity (DPPH: 22.00 $\mu\text{mol TE/g}$, ABTS: 31.33 $\mu\text{mol TE/g}$, FRAP: 11.24 mmol FeSO₄/g), in addition to the identification of 18 phenolic compounds, with procyanidin A2 being the major compound. The films with 1.5% GHRE showed better appearance, greater elasticity and lower permeability to water vapour (2.09×10^{-8} gH₂O.mm/m².h.MPa), which favours moisture-sensitive foods. The FTIR and XRD analyses revealed that the incorporation of GHRE promoted intermolecular interactions without altering the semicrystalline structure of the films. Thermal stability increased with GHRE addition, whereas film luminosity decreased. In addition, these films demonstrated accelerated biodegradability (> 96% in 30 days), indicating strong potential for sustainability applications. These results confirm the potential of GHRE as a functional additive in antioxidant and biodegradable active packaging.

1 | Introduction

The food industry has been seeking to develop new sustainable packaging materials, due to the serious environmental problems arising from the use of conventional packaging from fossil sources. Faced with this scenario, an alternative is the use of polymers from renewable, biodegradable, abundant and low-cost sources, such as biopolymers derived from polysaccharides, proteins and lipids [1].

In this context, biodegradable films based on proteins and polysaccharides have been widely investigated as ecological alternatives to conventional synthetic materials [2]. These films, produced as blends of polysaccharides and proteins,

demonstrated satisfactory performance in terms of mechanical strength, flexibility and efficiency as barriers against moisture, gases and volatile compounds [3, 4]. Additionally, recent work by Kurabetta et al. [5] showed that vanillin-reinforced cationic starch/PVA composite films achieved ~49% improvement in tensile strength and a ~60% reduction in water vapour permeability, demonstrating how phenolic modifiers can significantly boost both mechanical and barrier properties of biopolymer films.

Collagen, a structural protein of animal origin, stands out for its ability to form transparent, flexible and biocompatible films, with good affinity for other bioactive compounds. The primary structures of the peptide chains of collagens are built by repeating the sequence of amino acids. The incorporation of structural

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proteins can enhance certain physicochemical properties of the films, although it can compromise their tensile strength [3]. The performance of these films can be further optimized through structural modification strategies, such as chemical crosslinking and the choice of suitable solvents, resulting in cohesive and stable matrices [2]. Moreover, the embedding of nano-functional additives in biopolymer films is emerging; for example, Hunashyal et al. [6] developed chitosan-based active films embedded with neem-leaf-derived carbon dots which improved antioxidant functionality and extended shelf-life of prawns, pointing to the potential of nano-additive incorporation in active biodegradable films.

On the other hand, starch has great potential for film formation due to its gelling, oxygen barrier and continuous matrix formation properties. In addition to being widely available and low cost [7]. Sweet potatoes (*Ipomoea potatoes*) are a promising source of starch (80% of dry weight) with desirable functional characteristics, such as high amylose content (20%–30%), retrogradation capacity and thermal stability, in addition to being widely available in tropical and subtropical regions [8]. Sweet potato starch has gained attention for its potential in the production of biodegradable and edible films, offering a solution to reduce crop waste and provide eco-friendly packaging alternatives [9].

The incorporation of natural bioactive compounds in biodegradable matrices has allowed the development of active packaging, capable of acting in the preservation of food, extending its shelf life [3]. Among these compounds, phenolics stand out for their recognized antioxidant and antimicrobial properties, with the potential to inhibit lipid oxidation reactions and the growth of spoilage microorganisms. Although phenolics can be obtained from various plant sources, the use of agro-industrial residues as raw material for the extraction of these compounds has been an advantageous strategy from an economic, environmental and social point of view [10]. In fact, recent findings by Kurabetta et al. [11] highlight that phenolic cross-linkers and plant-derived antioxidants can enhance film strength, water resistance and bioactivity in polysaccharide systems, reinforcing the relevance of phenolic incorporation.

G. humilis, popularly known as achachairu, is a tropical species belonging to the *Clusiaceae* family, commonly cultivated in regions of South America due to its adaptability and promising nutritional profile [12]. Its fruit is consumed *in natura* and used in the production of pulps, juices and jellies, generating significant residues, especially peels and seeds, with a high content of phenolic compounds that are still little explored. In addition, its fruits are rich in vitamins, minerals and antioxidant compounds, standing out as natural sources of nutrients with functional applications [13]. The peel of the fruit contains a variety of bioactive compounds with potent antioxidant activity, which makes it an attractive ingredient for the fortification of foods and formulations with the potential to extend the shelf life of products [14]. Recent studies suggest that these residues can be rich sources of natural antioxidants, which could be used in controlled-release systems, cosmetics and, more recently, biodegradable matrices with food application. The multiple potential applications of achachairu, which span both the food and pharmaceutical sectors, reinforce the need for in-depth studies on its chemical, bioactive and technological properties [15].

Despite advances in research on films with functional properties, there is a significant gap in the literature regarding the synergistic combination of collagen and sweet potato starch with phenolic compounds from *G. humilis* residue extract (GHRE). Few studies have explored the full use of agro-industrial residues as active agents in food-compatible biopolymer systems. In addition, it remains unclear how this incorporation impacts the physicochemical, structural, antioxidant properties and biodegradability of the films generated [16].

In this context, the hypothesis of the study is that the addition of phenolic compounds extracted from *G. humilis* residues into collagen–sweet potato starch films will enhance their functional and structural performance. Specifically, GHRE is expected to (i) increase the antioxidant activity of the films due to its high content of phenolic acid and flavonoid compounds capable of scavenging free radicals; (ii) improve thermal stability by acting as a natural stabilizer that delays polymer degradation; (iii) modify mechanical and barrier properties through intermolecular interactions between extract constituents and the polymeric matrix, potentially increasing strength and reducing water vapour and gas permeability; and (iv) promote biodegradability owing to the organic nature of the extract, which may facilitate microbial degradation under natural conditions. Therefore, the present study aimed to develop and characterize active and biodegradable films composed of collagen and sweet potato starch incorporated with phenolic extracts obtained from *G. humilis* residues and to evaluate the effects of the extract on their antioxidant, thermal, mechanical, barrier and degradability properties.

2 | Materials and Methods

2.1 | Materials

Sweet potato starch (Empório Terra Vitta), collagen (Gelita do Brasil Ltda) and *G. humilis* (achachairu) fruits were purchased from local markets in João Pessoa, Paraíba, Brazil, and transported to the Federal University of Paraíba (UFPB), João Pessoa, PB, in insulated thermal containers.

The fruits were selected based on external appearance, ripeness stage and absence of physical damage. After selection, they were washed under running water, sanitized in a 200ppm sodium hypochlorite solution for 10 min and then manually pulped. The resulting peels and seeds were stored at -18°C until extraction and further analytical procedures.

The Folin–Ciocalteu reagent was supplied by Merck KGaA (Darmstadt, Germany). The reagents ferric chloride hexahydrate, sodium acetate trihydrate and sodium carbonate were acquired from VETEC (Duque de Caxias, RJ, Brazil). Gallic acid, 2,2-diphenyl-1-picrylhydrazyl, 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic (Trolox), methanol and acetonitrile, both HPLC grade, were purchased from Sigma-Aldrich Co. (St. Louis, MO, USA). The same company supplied 18 standards, namely, gallic acid, procyanidin B1, catechin, procyanidin B2, syringic acid, epicatechin, epigallocatechin gallate, myricetin, rutin, epicatechin gallate, quercetin, procyanidin A2, kaempferol, hesperidin, isorhamnetin, trans-resveratrol, cis-resveratrol

and naringenin. The other reagents and solvents of analytical grade, like ethanol and hydrochloric acid, were purchased from the company Química Moderna (Barueri, Paraíba, Brazil).

2.2 | Obtaining the *G. humilis* Residue Extract

The extraction was performed according to the procedure described by Adilah et al. [13], with minor modifications to improve reproducibility. All extractions were performed in triplicate. The *G. humilis* (achachairu) residue was dried in a forced-air oven at 60°C for 24 h and then ground to obtain a homogeneous powder. A total of 10 g of the powdered residue was homogenized with 100 mL of a 50% (v/v) ethanol–water solution, corresponding to a 1:10 solid-to-solvent ratio, and maintained under magnetic stirring (180 rpm) at 25°C for 4 h. After extraction, the mixture was filtered through standard filter paper, and the filtrate was concentrated under reduced pressure at 80°C using a rotary evaporator. The concentrated extract was then transferred to amber glass bottles with airtight lids and stored under refrigeration (5°C) until use.

2.3 | Determination and Identification of Phenolic Compounds and Antioxidant Activity of GHRE

2.3.1 | Determination of Total Phenolic Content

The phenolic compound concentration was determined using the method outlined by Singleton and Rossi [17]. A sample solution with an initial concentration of 5 mg/mL was prepared. For the assay, 150 µL of the sample was pipetted onto a glass tube, along with 60 µL of Folin–Ciocalteu reagent and 2610 µL of distilled water. After 1 min, 180 µL of sodium carbonate (15%) was added, the mixture was stirred for 1 min again, and it was left to stand for 2 h to react at (25°C). The absorbance readings of the samples and standard were measured at 760 nm using a spectrophotometer (Quimis, Model Q798U). A calibration curve was constructed using gallic acid standards ranging from 1 to 20 mg/L, prepared under the same conditions as the samples. Phenolic content was expressed as milligrams of gallic acid equivalents (GAEs) per gram of sample (mg GAE/g).

2.3.2 | Identification and Quantification of Phenolic Compounds

The profile of phenolic compounds was determined using the method described by Meireles et al. [18]. Chromatographic analyses were conducted on a Shimadzu High Performance Liquid Chromatograph (HPLC) system (Kyoto, Japan), equipped with a Rheodyne 7125i automatic injector and a UV/VIS detector. Separation was achieved using a Shimadzu LC-18 analytical column (25 cm × 4.6 mm, 5-µm particle size; Supelco, Bellefonte, PA), preceded by a C-18 ODS Shimadzu precolumn. Sample elution was carried out using a gradient mobile phase composed of solvent A (2% acetic acid, v/v) and solvent B (acetonitrile/methanol, 2:1, v/v) at a flow rate of 1.0 mL/min. The column temperature was maintained at 25°C, and the injection volume was

20 µL. The gradient program was as follows: 90% A at 0 min; 88% A at 3 min; 85% A at 6 min; 82% A at 10 min; 80% A at 12 min; 70% A at 15 min; 65% A at 20 min; 60% A at 25 min; 50% A from 30 to 40 min; 75% A at 42 min and 90% A at 44 min. The samples were eluted up to 50 min. Detection was performed at 280 nm. Stock standard solutions were prepared in methanol and subsequently diluted to obtain six to nine different concentration levels. Calibration curves were constructed from triplicate injections of each standard solution. Chromatogram acquisition and data processing were carried out using LabSolutions software (Shimadzu). Retention times were determined using external calibration curves constructed from the peak areas and concentrations of standard solutions, with regression coefficients (r^2) > 0.988. The identification and quantification of phenolic compounds in the extracts were performed by comparing the retention times and UV-Vis spectra of sample peaks with those of standards.

2.3.3 | 2,2-Asino-Bis (3-Ethylbezothiazoline)-6-Sulfonic Acid (ABTS•+)

The ABTS radical scavenging activity was assessed following the method described by Re et al. [19], with slight modifications. The ABTS radical cation was generated by reacting 5 mL of 7 mM ABTS stock solution (prepared in ethanol PA) with 88 µL of a 140 mM potassium persulfate solution (prepared in water). The resulting solution was then diluted with absolute ethanol at a 1:70 ratio to obtain the working solution. For the assay, 150 µL of the extract was mixed with 1000 µL of the diluted ABTS⁺ solution. After a 6 min incubation period at room temperature (25°C), the absorbance was measured at 734 nm using a spectrophotometer (Quimis, Model Q798U). Antioxidant activity was determined based on a Trolox standard curve (0–16 µM), and results were expressed as micromoles of Trolox equivalents per gram of sample (µmol TE/g).

2.3.4 | 2,2-Diphenyl-1-Picrylhydrazyl (DPPH•)

DPPH radical scavenging effect was measured following the method described by Brand-Williams et al. [20] with modifications. A mixture containing 150 µL of the extract and 150 µL of ethanol was homogenized with 2700 µL of 50 µM DPPH solution prepared in ethanol. The reaction mixture was kept in the dark at room temperature (25°C) for 30 min. After incubation, the absorbance was measured at 517 nm using a spectrophotometer (Quimis, Model Q798U). Antioxidant activity was calculated using a Trolox standard curve (0–2.5 µM), and the DPPH• radical scavenging capacity was expressed as micromoles of Trolox equivalents per gram of sample (µmol TE/g).

2.3.5 | Capacity by the Ferric Reducing Antioxidant Power Method

Antioxidant activity was assessed using the Ferric Reducing Antioxidant Power (FRAP) assay, based on the method proposed by Rufino et al. [21]. The FRAP reagent was freshly prepared by mixing acetate buffer (300 mM, pH 3.6), TPTZ solution (2,4,6-tripyridyl-s-triazine) solution (10 mmol/L in 40 mmol/L

HCl) and FeCl_3 solution (20 mmol/L) in a 10:1:1 (v/v/v) ratio. For the assay, 90 μL of the sample was combined with 270 μL of distilled water and 2700 μL of the FRAP reagent in test tubes. The mixture was homogenized and incubated in a water bath at 37°C for 30 min. Absorbance was then measured at 595 nm using a spectrophotometer (Quimis, Model Q798U), with a blank containing only the FRAP reagent. A standard curve was prepared using FeSO_4 solutions ranging from 50 to 2000 μM . Results were expressed as millimoles of FeSO_4 equivalents per gram of sample (mmol FeSO_4/g).

2.4 | Elaboration of the Films

The films were prepared by the method of *casting* according to the methodology described by Costa et al. [22]. The collagen-to-starch ratio was 1:2 on a dry basis. Collagen (2%, w/w) was dissolved in distilled water under stirring to obtain an aqueous solution, and the pH of the solution was adjusted to between 3 and 4 with citric acid. After that, sweet potato starch (4%, w/w) and the plasticizers glycerol (1.5%, w/w) and sucrose (0.7%, w/w) were added. Following manual homogenization for 2 min, the extract was incorporated at different concentrations previously reported as effective for bioactive film formulations. The following four formulations were prepared: C0 (0% GHRE, control), A1 (0.5% GHRE), A2 (1.0% GHRE) and A3 (1.5% GHRE) (supplementary material, Table S1).

The mixture was homogenized for another 10 min in a Turrax stirrer (IKA RW 20 digital) at room temperature (25°C). Then, the solution was heated to 70°C–75°C under constant stirring until starch gelatinization. Each film-forming solution (40 g, containing 2.38 g total solids) was poured into round acrylic Petri dishes (150 × 15 mm). The plates were placed in an air circulation oven at 40°C for 8–9 h. After drying, the films were conditioned at 25°C and 75% relative humidity for 48 h prior to analysis.

2.5 | Spectroscopic, Structural and Thermal Characterization

2.5.1 | Fourier Transform Infrared Spectroscopy

The infrared spectra of the films were obtained using Fourier Transform Infrared Spectroscopy with Attenuated Total Reflectance (ATR-FTIR), employing a Spectrum 8900 IR spectrometer (Shimadzu, Japan), as described by Kumar et al. [23]. Spectra were recorded over the wavenumber range of 4000–600 cm^{-1} at room temperature, with a resolution of 4 cm^{-1} and 40 scan accumulations per sample. Prior to measurement, the films were cut into small pieces to ensure proper contact with the ATR crystal. In addition to the film samples, ATR-FTIR spectra of the collagen and sweet potato starch used in the formulations were also collected to assist in identifying functional groups and monitoring interactions between components (supplementary material). Auto-baseline correction was applied to all spectra before curve fitting. Fourier self-deconvolution spectra were then generated with IR Solutions software (Shimadzu), and peak fitting was performed with OriginPro software, Version 8.5 (OriginLab Corporation, Northampton, MA, USA).

2.5.2 | X-Ray Diffraction

The crystalline structures of the polymeric components (collagen and sweet potato starch) as well as the resulting films were analyzed by x-ray diffraction (XRD) using a DX-1000 diffractometer (Dandong Fangyuan Instrument Co. Ltd.), following the methodology of Chen et al. [24]. Prior to analysis, the films were cut into squares of approximately 2 cm × 2 cm to fit the sample holder, ensuring a flat and uniform surface. Collagen and sweet potato starch powders were sieved through a 200-mesh screen to obtain uniform particle size and improve packing homogeneity during measurement (supplementary material). Measurements were performed at 25°C using Cu-K α radiation under operating conditions of 30 kV and 30 mA. Diffraction patterns were recorded over a 2 θ range of 5° to 60°, at a scanning rate of 1°/min. The obtained diffractograms were processed and analyzed using OriginPro software (Version 8.5, OriginLab Corporation, Northampton, MA, USA).

2.5.3 | Thermogravimetric Analysis and Differential Scanning Calorimetry

The thermal properties of the polymeric components (collagen and sweet potato starch, in supplementary material) and the resulting films were evaluated using thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC), according to the procedure of Batista et al. [25] with minor modifications. All film samples were cut into small pieces before weighing them to ensure uniform heating. TGA measurements were performed on a TGA-50 analyser (Shimadzu, Japan), using approximately 10 mg of sample placed in alumina crucibles under a synthetic air flow of 50 mL/min. The temperature increased from 25°C to 800°C at a heating rate of 10°C/min. DSC measurements were carried out on a DSC-60 calorimeter (Shimadzu, Japan), using approximately 7 mg of sample sealed in aluminium pans under a nitrogen atmosphere (50 mL/min). The temperature was ramped from 25°C to 550°C at a rate of 10°C/min. Thermograms, expressed as weight loss (%) versus temperature, and heat flow curves of the film and raw materials were analyzed using OriginPro 8.5 (OriginLab, Northampton, MA, USA).

2.5.4 | Instrumental Colour

The colour of the film (3 replicates per sample, $n=3$) was assessed using a colorimeter (Minolta, CR 300, Mahwah/New Jersey, USA), based on the CIELab system, where the L^* represents lightness, a^* corresponds to red colour (positive values) or green colour (negative values), and b^* indicates yellow colour (positive values) or blue colour (negative values). A standard white calibration plate was used to calibrate the colorimeter [26]. The colour difference (ΔE) was calculated relative to the control (C0) using Equation 1.

$$\Delta E = \left[(L^* - L)^2 + (a^* - a)^2 + (b^* - b)^2 \right]^{0.5} \quad (1)$$

2.5.5 | Solubility and Moisture

The determination of film solubility and moisture was performed based on the method proposed by Nor Amalini et al. [27] with slight modifications. Initially, film samples measuring 2 × 2 cm were accurately weighed and subjected to drying in an oven at 105°C for 24 h, and the moisture was calculated. Then, each sample was transferred to an Erlenmeyer flask containing 30 mL of distilled water maintained at 25°C. The mixture was kept under continuous stirring for 24 h. After the stirring period, the flask was placed back in the oven at 105°C for an additional 24 h. Upon completion of the drying process, the final weight of the remaining film residue was recorded. The water solubility of the films was then calculated using Equation 2:

$$\text{Solubility (\%)} = [(m_i - m_f) / m_i] \times 100 \quad (2)$$

where m_i indicates the initial mass of dry material (g) and m_f indicates the final mass that did not dissolve (g).

2.5.6 | Water Vapour Permeability

The water vapour permeability (WVP) tests were performed using the gravimetric method described in the ASTM E96/E96M-16 protocol [28]. Film samples were fixed within a properly sealed container (3 cm in diameter) containing 10 g of silica gel, positioned approximately 1.5 cm from the film. The initial mass of the system was measured and stored in a desiccator maintained at 25°C and 75% relative humidity. Over the course of 7 days, the system was weighed every 24 h in triplicate. The WVP was then calculated using the following equation:

$$\text{WVP} = [\Delta m / A \cdot \Delta t] \times (X / (P_s \times (RH1 - RH2))) \quad (3)$$

Where, WVP is water vapour permeability, $\Delta m / \Delta t$ is the slope of the weight gain curve per unit time, A is the exposed surface area of the film (m²), X is the average film thickness, P_s is the saturation pressure of the water, RH1 is the relative humidity inside the desiccator (75%), and RH2 is the relative humidity inside the capsule (0%). The results were expressed in gH₂O.mm/(m².h.MPa).

2.5.7 | Degradability

The degradability of the films was assessed by measuring mass loss resulting from exposure to the natural soil microbiota over a 30-day period, following the methodology described by Chandra and Rustgi [29]. Rectangular film samples (2 × 2 cm) were prepared, preconditioned at 25.00 ± 2.00°C and 70.00 ± 5.00% relative humidity for 48 h, weighed (initial mass, m_i), and buried at a depth of approximately 15 cm in plastic containers filled with natural soil collected from the experimental area of the Federal University of Paraíba (João Pessoa, Brazil). The soil exhibited a sandy-loam texture, pH 7.50, organic matter content of 5.20 ± 0.20% and moisture of 32.00 ± 1.00%. The containers were maintained at 25°C and 51.00 ± 3.00% of water-holding capacity (WHC) to ensure stable biological activity during the assay.

After 30 days, three samples per formulation ($n=3$) were carefully removed using sterile tweezers, rinsed with distilled water to eliminate adhering soil particles, and oven-dried at 40°C for 24 h until constant weight. The film without GHRE (C0) was used as the control for comparative evaluation of degradation. The percentage of degradation was calculated according to the following equation:

$$\text{Degradability (\%)} = [(m_i - m_f) / m_i] \times 100 \quad (4)$$

where m_i is the initial film mass (g) and m_f is the final dry film mass (g).

2.5.8 | Thickness and Mechanical Properties

To evaluate the mechanical properties, the average thickness of the films was determined from ten random measurements taken at positions located 60 mm from the edges, at room temperature (25°C). Measurements were performed using a portable micrometre with an accuracy of 0.001 mm. The results were expressed in millimetres (mm), and the mean value was used in subsequent calculations.

Tensile strength and elongation at break were determined using a static testing instrument (SHIMADZU, Japan), following the standard protocol described in ASTM D882-12 [30]. Film specimens were prepared in a rectangular format measuring 100 mm in length and 15 mm in width. The initial grip separation was set at 50 mm, and the crosshead speed was maintained at 12.5 mm/min during testing. Each treatment was analyzed in 10 replicates. The results were expressed in megapascals (MPa) for tensile strength and in percentage (%) for elongation at break.

2.6 | Statistical Analysis

Results were expressed as mean ± standard deviation and presented in tables and figures. Statistical comparisons among means and the identification of significant differences ($p < 0.05$) were performed using analysis of variance (ANOVA), followed by Tukey's test with the Assistat software (Version 7.7). Graphs were generated using Origin software (Version 8.0).

3 | Results and Discussion

3.1 | Determination and Identification of Phenolics and Antioxidant Activity of GHRE

The content of phenolic compounds of the GHRE obtained in the present study was 543.69 ± 2.37 mg GAE/g, higher than reported by Tome et al. [31] for the aqueous and ethanolic extracts of achachairu peels and seeds (160.50; 125.50 mg GAE/100 g, respectively). These results indicate that the hydroalcoholic extraction methodology employed was more effective in the extraction of bioactive phenolic compounds, probably due to the greater solubility of these compounds in solvents with intermediate polarity. The high concentration of phenolics observed in GHRE suggests the significant presence

of polyphenols, compounds recognized for their ability to act as free radical scavengers and iron chelators, as demonstrated in their antioxidant potential results [14].

The antioxidant activity of GHRE, evaluated by the DPPH methods ($22.00 \pm 0.23 \mu\text{mol TE/g}$), ABTS ($31.33 \pm 0.28 \mu\text{mol TE/g}$) and FRAP ($11.24 \pm 0.57 \text{ mmol FeSO}_4/\text{g}$), was also higher than that observed by Barros et al. [32] who used aqueous extracts obtained from the same plant residues. This performance reinforces the determining influence of the type and proportion of solvent on the efficiency of the extraction of antioxidant compounds. Phenolic compounds, widely recognized as the main responsible for the neutralization of free radicals and the reducing capacity in biological systems, have polarity variations that directly impact their solubility and extraction [33]. In this context, Gomes et al. [14] demonstrated that the use of hydroalcoholic extraction in the ratio of 90:10 (ethanol:water) was particularly effective in the extraction of phenolic compounds and high antioxidant activity. These findings corroborate the results obtained in the present study, suggesting that the appropriate choice of solvent system, especially mixed solvents with adjusted polarity, is crucial to maximize the yield of bioactive compounds and, consequently, the antioxidant potential of the extracts [34].

ABTS radical inhibition was superior to DPPH radical inhibition, indicating greater affinity of the compounds present in the extract for mechanisms based on electron and hydrogen transfer, being more versatile and sensitive to a wide range of hydrophilic and lipophilic antioxidants. This behaviour is typical of phenolic antioxidants with high cationic free radical stabilization capacity soluble in both aqueous and organic media [35]. On the other hand, the less intense response in the FRAP assay suggests a lower concentration of compounds acting predominantly by electron transfer in an acidic medium, a mechanism evaluated in this specific test [36].

The antioxidant values in this study are consistent with literature reporting that hybrid organic-inorganic or phenolic-enriched films can deliver significant radical-scavenging activity while also contributing to the functional stabilization of the matrix. For example, films incorporating metal oxide nanoparticles into chitosan/gelatin or phenolic-crosslinked polysaccharide matrices have shown improved oxidative stability of packaged produce and enhanced preservation outcomes [37]. Moreover, the role of nano- or phenolic-mediated interactions in altering polymer crystallinity and thermal behaviour has been demonstrated by Pinto et al. [38], providing mechanistic support to the antioxidant-stabilizing effects observed here.

For phenolic and flavonoid profiles, 18 compounds were identified, whose concentrations are shown in Table 1. Procyanidin A2, hesperidin, rutin, myricetin, epicatechin gallate and naringenin were the major compounds, especially procyanidin A2, with a concentration of 110.783 mg/L. The isomers epicatechin and procyanidin were also identified in previous studies of extracts from achachairu bark. Extracts containing these isomers are known to exhibit antioxidant activities [39]. Numerous studies have reported that flavonoids exhibit diverse biological benefits [40]. In the studies carried out by

TABLE 1 | Concentrations of GHRE phenolic compounds.

Phenolic compounds		
Compound	Retention time (min)	Concentration (mg/L)
Gallic acid	7.40	1.71
Procyanidin B1	12.75	0.29
Catechin	14.61	2.04
Procyanidin B2	16.68	4.01
Syringic acid	17.31	4.05
Epicatechin	19.14	1.49
Epigallocatechin gallate	19.70	5.56
Myricetin	24.01	35.06
Rutin	25.10	42.51
Epicatechin gallate	25.43	16.89
Quercetin	25.74	0.95
Procyanidin A2	26.05	110.78
Kaempferol	26.92	3.14
Hesperidin	27.30	47.95
Isortohamnetine	27.61	0.54
Trans-resveratrol	29.38	0.45
Cis-resveratrol	31.97	1.20
Naringenin	33.67	10.18

Barros et al. [32], only eight phenolic compounds were identified in lower concentrations, evaluating aqueous and methanolic extracts of achachairu residues (epicatechin: 0.09601 mg/g; ferulic acid: 0.09601 mg/g). However, according to the literature, factors such as sample preparation, polarity of the solvent used, technique employed and temperature can interfere in the extraction and consequently in the quantification of these compounds in extracts [41].

The findings of the present study align with the current understanding that phenolic compounds enhance the functional performance of active packaging films by providing radical-scavenging activity that delays oxidative deterioration of food products. The mechanism is associated with the ability of hydroxyl groups in polyphenols to donate hydrogen atoms, forming stable structures that quench free radicals such as DPPH• and ABTS•+. Recent works have emphasized the importance of integrating such natural antioxidants into biodegradable matrices to prevent oxidation-induced spoilage, off-flavour development and nutrient loss during food storage [11].

Therefore, the strong antioxidant potential of GHRE reinforces its applicability as a natural active additive for food-packaging materials, providing protective activity against oxidative degradation and extending product shelf life through controlled radical scavenging mechanisms [42].

3.2 | Films Characterization

3.2.1 | FTIR Spectroscopy

The ATR-FTIR spectra of the collagen–starch films, both without (C0) and with GHRE incorporation (A1–A3), are presented in Figure 1. All film formulations exhibited characteristic absorption bands associated with the protein (collagen) and polysaccharide (starch) constituents, indicating the successful integration of both biopolymers in the matrix.

The broad band observed around 3329cm^{-1} corresponds to O–H and N–H stretching vibrations, and its intensity decreased with rising GHRE content (A1–A3). This shift and broadening suggest the formation of new hydrogen bonds between the hydroxyl groups of the phenolic compounds present in GHRE and the polar sites in the film matrix, consistent with phenolic–protein/polysaccharide interactions reported in the literature [43].

The peak at 2912cm^{-1} relates to C–H stretching vibrations, commonly attributed to aliphatic groups. This band became slightly more pronounced in GHRE-containing films, possibly due to the incorporation of hydrophobic components from the extract. While in the fingerprint region, three well-defined amide-related peaks were observed: amides at 1649cm^{-1} (C=O stretching of peptide bonds), at 1411cm^{-1} (N–H bending/C–N stretching) and at 1037cm^{-1} (C–N stretching and N–H deformation). The amide peaks showed minor shifts and increased intensity in films with GHRE, which may indicate enhanced molecular interactions and structural rearrangement due to the presence of phenolic compounds. Additionally, the increased definition of the 1037cm^{-1} band in A2 and A3 suggests a higher degree of hydrogen bonding and matrix organization, which could contribute to improved film cohesion, thermal and mechanical properties [44].

Overall, the similarities between the spectra of pure polymeric matrix components collagen and starch (Supplementary Material, Figure S1) and the films' spectra (Figure 1) confirm that the structural backbone of the films is maintained through

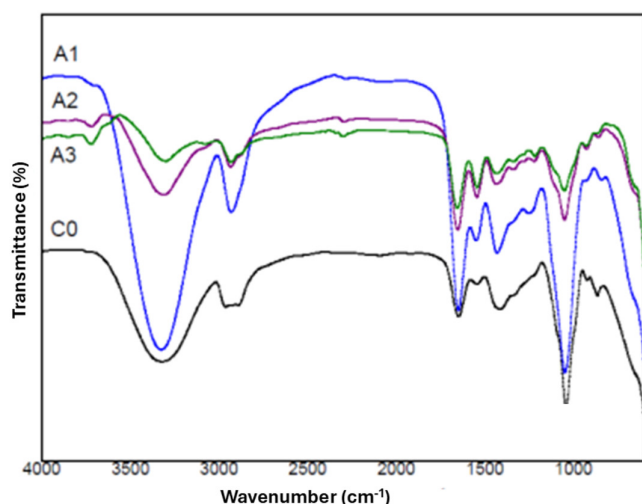


FIGURE 1 | ATR-FTIR spectra of collagen–starch-based films: control film without GHRE (C0) and films incorporated with GHRE at 0.5% (A1), 1.0% (A2) and 1.5% (A3).

characteristic functional groups of starch and collagen. Thus, the incorporation of GHRE into the biopolymeric matrix does not disrupt the primary functional groups of collagen and starch, but instead promotes intermolecular interactions, particularly through hydrogen bonding. These interactions may be key to the enhanced physicochemical properties observed in GHRE-enriched films.

3.2.2 | X-Ray Diffraction

The XRD patterns of the composite films (C0, A1, A2, A3) are shown in Figure 2. For comparative purposes, the XRD spectra of the individual biopolymeric components (sweet potato starch and collagen) are provided in the Supplementary Material (Figure S2). All film formulations exhibited a broad halo between 16° and 22° 2θ , indicating a semicrystalline structure, consistent with the combination of crystalline starch and amorphous collagen.

The control film (C0) presented two main diffraction peaks at 16.92° and 19.50° , with a shoulder near 21.80° , reflecting partial recrystallization of starch (primarily amylose) during film formation. The hydrothermal treatment applied during gelatinization disrupts the native crystalline structure of starch, converting it into an amorphous state [45]. However, upon cooling and drying, amylose chains tend to recrystallize through intermolecular hydrogen bonding, especially under high water mobility, as reported by Issa et al. [46] and Song et al. [47]

This recrystallization is further influenced by amylopectin, which may undergo a slower, humidity-dependent recrystallization during film storage or drying. Though co-crystallization between amylose and amylopectin may occur, it is generally limited. This behaviour may explain the observed semicrystalline pattern in C0, with collagen contributing predominantly amorphous character to the matrix [48].

For the GHRE-containing films (A1, A2, A3), no new peaks were observed compared to C0. Slight shifts in peak positions

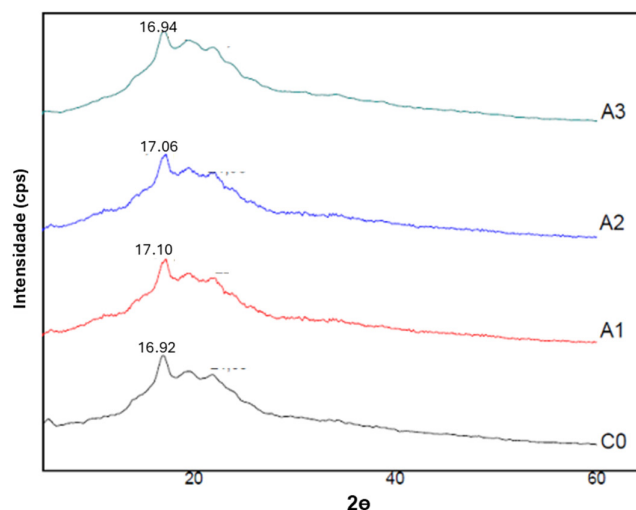


FIGURE 2 | XRD patterns of collagen–starch-based films: control film (C0) and films incorporated with GHRE at 0.5% (A1), 1.0% (A2) and 1.5% (A3).

(e.g., to $\sim 17.10^\circ$, 19.60° , 22.04°) and similar relative intensities suggest that the incorporation of GHRE did not significantly disrupt or enhance crystalline order. The lack of additional crystalline domains implies amorphous dispersion of GHRE within the matrix. Similar observations were reported by Nur Amila Najwa et al. [49] for gelatin–cassava starch films with phenolic extracts.

In summary, the XRD analysis suggests that the semi-crystalline nature of the films originates from partial amylose and amylopectin recrystallization, influenced by film processing conditions. GHRE presence may interact with matrix components, but at the concentrations tested, it does not significantly modify the crystalline organization of the films.

3.2.3 | Thermogravimetric Analysis

The thermogravimetric (TGA) and derivative thermogravimetric (DTG) curves of the films are presented in Figure 3, and their respective degradation temperatures and weight losses (Δm) are summarized in supplementary material, Table S2. All film samples exhibited a typical three-stage thermal degradation behaviour.

The first weight loss stage, observed between approximately 55 and 70°C , corresponds to the evaporation of physically adsorbed water and moisture. Notably, the control film (C0) exhibited the lowest mass loss in this range, suggesting lower moisture content or more tightly bound water, likely due to the absence of GHRE, which may introduce more hydrophilic sites.

The second stage, occurring roughly between 160 and 260°C , is attributed to the decomposition of glycerol, used as a plasticizer. This is consistent with the reported thermal degradation temperature of glycerol around 182°C [50]. Minor variations across

samples in this stage suggest an interaction between GHRE concentration and matrix plasticization.

The third and most significant mass loss stage, extending from approximately 250 to 490°C , corresponds to the degradation of collagen-rich protein fractions [51]. All samples exhibited the highest mass loss in this region, indicating that collagen is the major thermally labile component at elevated temperatures.

Among the samples, C0 and A3 showed greater thermal stability, as evidenced by their higher residual mass values (13.67% and 8.37%, respectively), suggesting enhanced carbonaceous residue formation. Interestingly, a trend of increasing residual mass with higher GHRE content (A1: 0.58%, A2: 4.44%, A3: 8.37%) was observed, indicating that GHRE contributes positively to thermal stability, possibly through polyphenol–protein or polyphenol–polysaccharide interactions that hinder full degradation.

Similar trends were reported by Costa et al. [22] in collagen/cassava starch films and by Ren et al. [52] in chitosan–collagen films incorporated with red cabbage extract. Tang et al. [53] also reported enhanced residual mass in collagen-based films modified with oxidized phenolic acids, reinforcing the stabilizing effect of polyphenolic components.

3.2.4 | Differential Scanning Calorimetry

DSC analysis revealed a broad endothermic peak around 120°C in all formulations (Figure 4), corresponding to the evaporation of absorbed water and starch gelatinization, as reported by Zaidul et al. [54]. Although protein denaturation events are typically observed below 100°C [55], such transitions were not clearly detected in either the collagen or film thermograms. This result aligns with

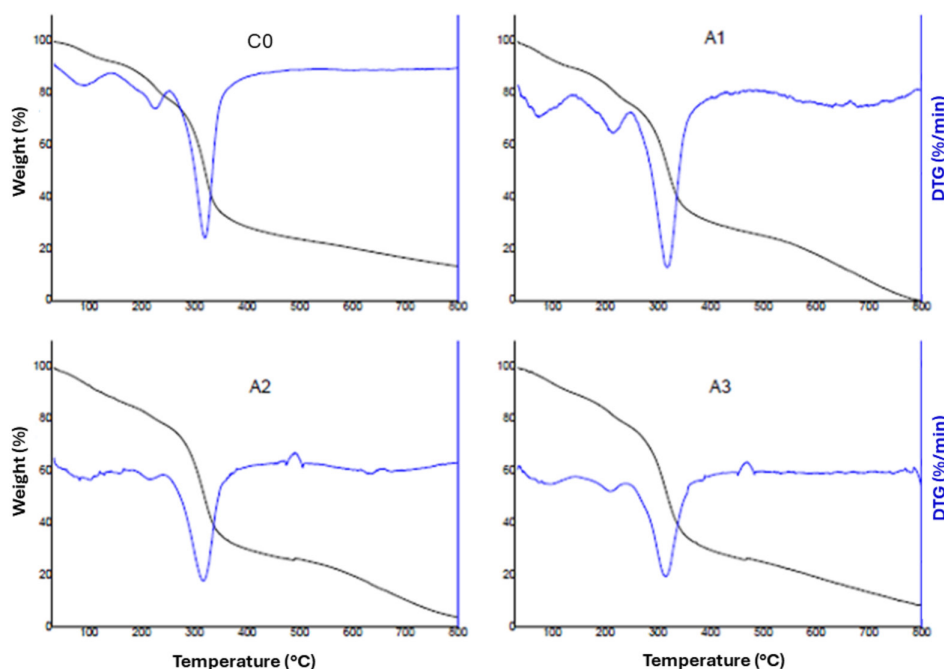


FIGURE 3 | TGA and DTG curves of composite films based on collagen, sweet potato starch and GHRE: C0 (0.0% GHRE), A1 (0.5% GHRE), A2 (1.0% GHRE) and A3 (1.5% GHRE).

the DSC profiles of the isolated components presented in the supplementary material (Figure S3), confirming the inherent thermal stability of the individual biopolymers. The absence of distinct denaturation peaks suggests that the collagen incorporated into the films exhibits enhanced structural resilience, thereby contributing to the improved thermal stability of the composite matrix.

In addition, the control film (C0), which contained no GHRE, showed signs of thermal degradation at lower temperatures compared to the films containing GHRE (A1, A2, A3). A progressive increase in thermal stability was observed with increasing GHRE content, indicating that the extract contributes to thermal protection of the matrix. This behaviour may result from interactions between polyphenolic compounds in the extract and the biopolymeric network, which stabilize the system. Similar findings were reported by Subbuvel and Kavan [56], who noted that curcumin enhanced the thermal stability of PLA films, although other additives such as fenugreek essential oil had a destabilizing effect.

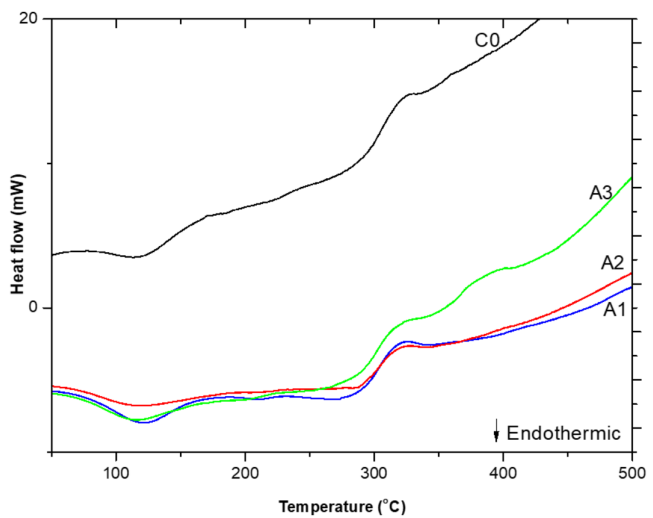


FIGURE 4 | DSC thermograms of collagen-starch-GHRE films: C0 (0.0% GHRE), A1 (0.5% GHRE), A2 (1.0% GHRE) and A3 (1.5% GHRE).

These observations are consistent with the TGA/DTG results, which demonstrated higher residual mass values for GHRE-containing films, particularly A3, supporting the conclusion that GHRE contributes to improved thermal stability. The absence of collagen denaturation peaks in the DSC, combined with the enhanced degradation resistance observed in TGA/DTG, suggests that phenolic compounds from GHRE may engage in stabilizing interactions with the protein and starch components, reinforcing the biopolymeric matrix.

3.3 | Optical and Physicochemical Properties of Films

3.3.1 | Colour

The appearance of the films is important to consider before using them as active packaging. The evaluation of the colour parameters for collagen, sweet potato starch and GHRE films is presented in Table 2.

The concentration of GHRE significantly affected all parameters ($p < 0.05$). For luminosity (L^*), the C0 film presented a higher value, and the higher the extract concentration, the lower the L^* value of the other films A1, A2 and A3 ($p < 0.05$).

Regarding the values a^* and b^* as well as ΔE , the A3 film presented the highest value ($p < 0.05$), with the red and yellow colours prevailing when compared to the other films, and the greatest colour difference in relation to the C0 film. In contrast to the L^* value, the addition of GHRE was accompanied by a significant increase in these parameters. This colour modification can be attributed to the nature of the extracts used. Plant extracts, which are potent sources of polyphenols, promote different types of interactions with biopolymers, including hydrogen bonds, hydrophobic interactions and complexation with matrix functional groups, which result in changes in the structural and optical properties of the final material [57].

TABLE 2 | Chromaticity values and images of collagen, sweet potato starch and GHRE films.

Formulations	L^*	a^*	b^*	ΔE	Images
A1 (0.5%)	90.41 ± 0.01^b	0.37 ± 0.02^b	11.46 ± 0.07^c	7.28 ± 0.04^c	
A2 (1%)	89.42 ± 0.01^c	0.20 ± 0.02^c	13.81 ± 0.04^b	9.80 ± 0.04^b	
A3 (1.5%)	87.51 ± 0.04^d	0.80 ± 0.02^a	16.99 ± 0.03^a	13.54 ± 0.04^a	
C0 (0%)	93.61 ± 0.01^a	-0.65 ± 0.02^d	4.99 ± 0.02^d	NA	

Note: The values are represented as mean $\pm$ standard deviation. Different letters in the same column indicate significant differences ($p < 0.05$) by Tukey's test. Abbreviation: NA, not applicable.

The predominance of red and yellow colours in the A3 film may be associated with the coloration of the GHRE and its higher concentration in this film. Among the residues, the peel is predominant (47% of the weight of the fruit) and has an orange-brownish colour when the fruit is ripe [58], stage in which the fruit was acquired to obtain the GHRE. Jiang et al. [59] found similar values for L^* (89.19 to 92.34) and lower values for parameters a^* (−1.27 to −1.8), b^* (2.17 to 3.51) and ΔE (0.39 to 1.77) of films based on carp collagen, chitosan and lemon essential oil. In addition, the parameters b^* and ΔE showed the same behaviour as in this study. These results are in line with those described by Jiang et al. [59] and Moghadam et al. [60] who observed similar increases in colour parameters (a^* , b^* and ΔE) in polymeric films with natural compound additives. Similar results were also found by Shao et al. [61] when analysing bovine bone collagen and hydroxypropyl methylcellulose (HPMC) films added with titanium dioxide [62].

The visual aspect of the packaging, particularly its colour and luminosity, plays a crucial role in the acceptance of the product by the consumer, influencing both its commercial attractiveness and its functionality. Films with greater transparency and less colour variation tend to be preferred in the market, as they allow a realistic visualization of the packaged food. However, this preference can be reversed in contexts where the protection of food against light is necessary. Photosensitive food products require packaging with lower light transmittance, capable of attenuating unwanted photo-oxidative reactions [57]. In this sense, the progressive darkening and increased opacity observed in films with a higher concentration of GHRE may represent a technological advantage, adding optical barrier properties to the packaging material.

3.3.2 | Moisture Content, Solubility, WVP and Degradability

The films without the addition of GHRE (C0) had the lowest moisture content, while the films with the highest concentration of GHRE (A3) had the highest content, showing that the presence and amount of extract added to the films directly influenced this parameter (Table 3). In addition to presenting the highest moisture content (17.26% and 18.30%, respectively), the films with the highest concentrations of GHRE (A2 and A3)

had the highest percentages of mass loss after 30 days of burial (96.63% and 96.75%, respectively). This shows that moisture facilitated the metabolic activities of soil organisms on the films, accelerating their degradation.

These results suggest that the increase in moisture content promoted the acceleration of the film degradation process, which agrees with previous studies that indicate the moderate humidity conditions (about 60%) favour the more efficient decomposition of biodegradable films. In addition, greater water availability can stimulate the activity of specific microbial communities, leading to the degradation of polymeric materials [51]. These microorganisms, under favourable environmental conditions, intensify film degradation by increasing the expression of enzymes and metabolic pathways related to the breakdown of organic compounds [63]. Film moisture control is a fundamental parameter to be monitored during film production, as it directly influences the shelf life of both the material and the food packaged [64].

Regarding solubility, the films with higher concentrations of GHRE showed lower contents, a result that can be attributed to the presence of hydrophobic phenolic compounds present in the extract, such as gallic acid, catechin, myricetin, quercetin and kaempferol, which have hydrophobic aromatic structures that limit their affinity with the aqueous matrix. Previous studies demonstrate that the presence of these compounds can reduce the water solubility of biopolymeric films, as noted by Bhatia et al. [65] who reported reduced solubility and moisture content of gelatin-casein films cross-linked with gallic acid. Similarly, Zhang et al. [66] showed that the incorporation of flavanols such as myricetin, quercetin and kaempferol into chitosan films significantly altered their barrier properties and interaction with water, reinforcing the influence of the hydrophobic nature of these compounds on the modulation of the physicochemical characteristics of the films.

Packaging with reduced WVP, consequently, is a packaging with improved water vapour barrier property between the food and the external environment, prolonging the characteristics and quality of food with this specific need. The WVP quantifies the rate at which water vapour passes through a polymeric material and is a fundamental parameter for evaluating the film's ability to act as a moisture barrier. A lower

TABLE 3 | Moisture content, solubility, water vapour permeability and degradability of the films.

Parameters	A1 (0.5%)	A2 (1%)	A3 (1.5%)	C0 (0%)
Moisture (%)	15.42 ± 0.43 ^{ab}	17.26 ± 1.37 ^{ab}	18.30 ± 1.92 ^a	14.78 ± 0.30 ^b
Solubility (%)	22.50 ± 3.53 ^{ab}	28.15 ± 2.09 ^a	9.47 ± 1.81 ^b	15.68 ± 7.21 ^{ab}
WVP*	6.28 ± 0.14 ^b	7.66 ± 0.28 ^a	2.09 ± 0.70 ^c	1.46 ± 0.21 ^c
Degradability (%)	41.33 ± 3.64 ^b	96.63 ± 3.36 ^a	96.75 ± 3.42 ^a	81.14 ± 13.55 ^a
Thickness (mm)	0.14 ± 0.01 ^b	0.14 ± 0.01 ^b	0.17 ± 0.01 ^a	0.11 ± 0.01 ^b
RT (MPa)	11.14 ± 1.98 ^a	9.30 ± 0.47 ^a	6.13 ± 0.35 ^b	10.59 ± 2.18 ^a
Elongation (%)	15.67 ± 3.62 ^b	14.86 ± 1.12 ^b	22.67 ± 2.64 ^a	13.89 ± 0.59 ^b

Note: The values are represented as mean ± standard deviation. Different letters on the same line indicate significant differences ($p < 0.05$) by Tukey's test.

*WVP—water vapour permeability in 10^{-8} gH₂O.mm/(m².h.MPa).

WVP indicates better resistance to water vapour diffusion, meaning that the material can more effectively prevent moisture penetration, which could otherwise cause degradation, oxidation or spoilage of packaged foods. Therefore, WVP is a critical property in food packaging applications, where maintaining optimal humidity levels is essential to preserve product quality and extend shelf life [11, 63].

A2 film showed higher WVP followed by A1 film ($p < 0.05$). On the other hand, the films with the highest concentration of GHRE (A3) and those without extract (C0) had the lowest values. The lower WVP values observed in A3 and C0 films suggest improved barrier performance, consistent with enhanced network cohesion and reduced free volume within the matrix. In the control film (C0), the dense collagen–starch structure likely limited water molecule diffusion pathways, while in the GHRE-containing film (A3), hydrogen bonding between phenolic hydroxyl groups and polymer chains may have filled microvoids, increasing matrix compactness and hindering vapour permeation [44]. The lower WVP values may be due to the higher electrostatic interactions between the collagen and starch (C0) biopolymers [67, 68], and due to the electrostatic compatibility of GHRE with the biopolymer matrix when added in specific concentrations such as 1.5% (A3), where the functional groups of phenolics are dispersed uniformly along the collagen–starch structure, reducing its ability to make hydrogen bonds with the medium [50].

Similar results have been reported in the literature, indicating a reduction in WVP in starch-based active films containing phenolic compounds. This decrease was attributed to the uniform dispersion of active molecules within the polysaccharide matrix, which reduced interstitial voids and blocked the pathways for vapour transmission [5].

These results are in line with previous studies reporting that the addition of natural extracts can, depending on the concentration and compatibility with the matrix, significantly alter the barrier properties of the films. Films with higher concentrations of natural additives, such as plant extracts, generally have higher WVP compared to control, due to increased polarity or structural heterogeneity of the matrix [69]. However, when incorporated in appropriate concentrations, these extracts can promote intermolecular interactions that improve the compactness and stability of the film structure [70], which may explain the lower WVP observed in the A3 film. This indicates that an optimal level of phenolic extract contributes to denser polymer packing and stronger intermolecular interactions, which reduce water diffusion through the film structure. A comparable nonlinear effect was observed by Eelager et al. [71], who evaluated PVA/xanthan gum films crosslinked with cetrimide. Their results revealed that crosslinking density and additive concentration markedly influenced film compactness, permeability and degradation behaviour.

For degradability, the films with the highest concentrations of GHRE (A2 and A3) and those without additives (C0) achieved mass loss percentages of almost 100% (96.63% and 96.75%) and 81.14%, respectively, higher than that of the A1 film (41.33%) during 30 days of burial. This shows that the addition of GHRE by 1% accelerates the degradation of films. Chemical

composition, material thickness and environmental conditions influence degradation rates of degradable films [72, 73].

Starch-based films, for example, demonstrate high biodegradation potential, with commercial formulations such as Mater-Bi showing mass losses of up to $45.0 \pm 4.7\%$ under simulated composting conditions [74]. Modification of the chemical composition, such as the addition of hydrolyzed collagen to alginate polymer matrices, also directly affects the performance and degradation behaviour of materials [75]. In addition, environmental factors such as temperature, humidity and soil microbial activity are determinants of the rate of film degradation. The thickness of the material, in turn, influences surface exposure and the diffusion of degrading agents, being especially relevant in materials such as polylactic acid (PLA) [76]. Thus, an integrated understanding of these parameters is essential for the development of biodegradable packaging with environmentally friendly performance [77].

The thickness and mechanical properties of collagen, sweet potato starch and GHRE films are presented in Table 3. The increase in the concentration of GHRE resulted in thicker films, a behaviour that can be attributed to the increase in the viscosity of the filmogenic solution, promoted by the intermolecular interactions between the bioactive compounds present in the extract and the biopolymers of the matrix. These compounds, rich in polar functional groups, can interact with polymeric chains through hydrogen bonds, contributing to the formation of a more cohesive network structure and, consequently, to increasing thickness during the drying process [78]. Chou et al. [79] reported lower thicknesses for fish skin gelatin-based films embedded with guava leaf extract, ranging from 0.053 to 0.068 mm. The authors observed that the films containing the extract exhibited greater thickness, which increased proportionally with the extract concentration. This behaviour corroborates the results obtained in the present study and agrees with the findings reported by Fernandes et al. [80]. Similarly, recent studies have demonstrated that the incorporation of bioactive extracts or nanofillers can markedly influence film uniformity and morphology by affecting polymer chain arrangement and solid content. Increasing the concentration of bioactive additives commonly enhances solution viscosity and solid density, resulting in thicker and smoother films with improved interfacial adhesion to substrates [81].

Regarding the mechanical properties, an inverse relationship was observed between tensile strength and elongation at break. With the increase in the concentration of GHRE, the films showed lower mechanical resistance and greater deformation capacity. This behaviour can be explained by the interference of the compounds of the extract in the structural organization of the polymeric matrix, reducing the interactions between the chains and promoting greater molecular mobility. Similar trends have been described for other polymeric systems, such as chitosan–polyvinyl alcohol, where the incorporation of natural agents modified the stress–strain behaviour [5].

This effect may be related to the low compatibility between the polymers in the matrix and the compounds present in the extract, many of which have complex spatial structures. This mismatch can result in a less compact structure, favouring the

formation of more flexible films, but with less structural cohesion, which compromises their tensile strength [57]. Pająk, Przetaczek-Rożnowska and Juszczak [82] reported tensile strength values comparable to those obtained in the present study, ranging from 8.98 to 13.85 MPa, as well as lower percentages of elongation at break (3.35%–4.44%) in films made with starches from unconventional sources, such as pumpkin fruits, lentils and quinoa seeds.

However, at moderate extract concentrations, hydrogen bonding between GHRE phenolics and polymer hydroxyl groups may also create a partially cross-linked network, which temporarily enhances tensile strength by restricting molecular motion. This dual behaviour, reinforcement at moderate concentrations and softening at higher levels, has been widely observed in bioactive-filled films, where the balance between crosslinking density and phase compatibility dictates the final mechanical response [83]. At higher GHRE loadings, the decrease in TS can be further attributed to phase heterogeneity and limited miscibility between the polymeric matrix and extract components. The formation of localized aggregates or microdomains can act as stress concentrators, disrupting force transmission through the matrix [84].

These results indicate that GHRE incorporation modified the stress–strain behaviour of the films through a complex interplay of hydrogen bonding, partial crosslinking and phase dispersion. While the films became more flexible and extensible, their tensile resistance declined beyond the optimal concentration, reflecting a typical trade-off observed in natural bioactive–reinforced systems. These findings align with literature trends for starch and protein-based biodegradable films, confirming that controlled incorporation of GHRE can fine-tune the mechanical balance between rigidity and flexibility, which is essential for tailoring packaging materials to specific applications.

4 | Conclusions

The results demonstrated that the incorporation of GHRE in collagen and starch matrices of sweet potato promotes significant improvements in the antioxidant, thermal, optical, mechanical and barrier properties of the developed films. In particular, the formulation with 1.5% GHRE stood out for its elasticity, thermal stability, antioxidant action, as well as high biodegradability. These findings position the films as promising active packaging, capable of replacing conventional plastic materials, especially in contexts that require resistance to moisture and oxidation, reinforcing their viability as an ecological and functional alternative in the food industry.

Despite the promising functional properties of GHRE-incorporated films, some limitations must be acknowledged. The current study focused on laboratory-scale casting, and scalability to pilot or industrial production may require optimization of drying rates, viscosity control and surface uniformity to ensure reproducibility. In addition, although the materials used are naturally derived and generally recognized as safe, specific evaluations of food-contact safety and potential migration of active compounds should be conducted in future work. Further studies should also explore the applicability of GHRE in other biopolymer matrices to evaluate its versatility and potential for

broader industrial use. These directions will help to translate the findings of this study into practical applications for sustainable and functional food packaging systems.

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Consent

All authors of this research work participated directly in the planning, execution or analysis of this study. All authors of this article have read and approved the final version submitted. All authors concur with the submission in Packaging Technology and Science.

Data Availability Statement

Data will be made available upon request.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Table S1:** Composition of films formulations. **Figure S1:** ATR-FTIR spectra of the polymeric matrix components: sweet potato starch and collagen. **Figure S2:** XRD pattern of the polymeric matrix components: sweet potato starch and collagen. **Table S2:** Thermal profile of collagen, sweet potato starch and GHRE-based films. **Figure S3:** DSC thermograms of isolated biopolymeric matrix components: sweet potato starch and collagen.