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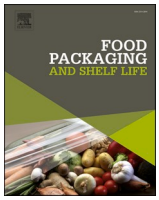

Bioactive PLA packaging films using artichoke leaf extract as functional additive

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ABSTRACT

Valorizing agro-food side streams to develop active packaging materials is a promising circular approach that helps reintegrate these residues back into the food supply chain. Herein, artichoke leaves were used as a source of bioactive extracts for the formulation of active PLA-packaging materials. The incorporation of artichoke leaves extract (ALE) in PLA-based films yielded films with no prominent structural or water vapor barrier changes compared to neat PLA films. Regarding mechanical properties, ALE contributed to the formation of films with less stretchability without affecting their stiffness. On the other hand, the ALE-incorporated films presented good UV shielding, and improved antioxidant activity compared with pure PLA films. Moreover, global migration assays revealed the different behavior of PLA-ALE films according to the food simulant, where the highly polar solvent (10 % ethanol) represented the most promising medium for the developed materials, allowing to establish the safety and regulatory compliance of up to PLA-ALE2.5. Thus, the developed PLA-ALE active materials, besides showing potential for foodstuff preservation, can also be considered safe for packaging applications.

1. Introduction

Agro-industrial operations are responsible for large food losses from field to processing, resulting in significant economic losses and resource depletion. Worldwide, about 13.2 % of food produced for human consumption is lost after harvest and before reaching retail, which adds to the 19 % of food wasted at the retail, food service, and household levels (FAO, 2022; UNEP, 2024). This scenario underscores the need for a sustainable food chain and a shift from the linear model to a circular economy by redefining waste and promoting conversion of side-streams into valuable products. Among the different agro-food products generating high waste, artichoke (*Cynara cardunculus var. scolymus* L.) discards 60–85 % of inedible parts (Zayed & Farag, 2020). Global production reached approximately 1.6 million tons (FAOSTAT, 2023), highlighting its impact on the food chain, in this way, solutions to make the artichoke chain more sustainable are needed. Moreover, artichoke byproducts are rich in phenolic compounds such as caffeic acid derivatives, cynarin, luteolin, and apigenin (López-Salas et al., 2021), offering opportunities for valorization as natural additives, e.g., in food packaging. Among extraction technologies, subcritical water extraction (SWE) has shown great potential for recovering bioactive compounds from agro-food byproducts without the use of organic solvents and with high efficiency in shorter times (Zhang et al., 2020). SWE-derived extracts have been successfully applied in active packaging, such as gelatin films with coffee ground extracts (Getachew et al., 2021) and PLA films with rice straw extracts, which preserved meat quality by reducing oxidation, discoloration, and weight loss (Freitas et al., 2023). Furthermore, sequential SWE (s-SWE) enables fractionation of compounds based on solubility, broadening the range of recovered bioactive compounds and demonstrating its versatility in biorefinery applications (Rincón et al., 2021).

Poly(lactic acid) (PLA), a bio-based polyester synthesized from lactic acid derived from agro-resources such as sugarcane, potato, and corn, has emerged as a promising alternative to conventional plastics (Swetha et al., 2023). Its transparency, non-toxicity, thermoprocessability, biodegradability under controlled composting conditions, and biocompatibility make it suitable for food packaging applications (Guicherd et al., 2024). Recent studies have focused on enhancing PLA

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functionality by incorporating natural bioactive compounds such as green tea and rice straw, which improve its antioxidant and antimicrobial properties and extend food shelf life (Martins et al., 2018; Freitas et al., 2023).

Previous research has demonstrated the improved biological potential of chitosan incorporated with ethanolic extracts of artichoke leaf as wound dressing materials (Brás et al., 2020). Similarly, Grimaldi et al. (2022) developed active alginate films with artichoke and onion byproduct extracts, enhancing meat and fruit preservation. However, deeper understanding of artichoke byproduct extracts' impact on packaging structure and bioactivity is still required. In this context, the aim of this research was to explore a valorization pathway for artichoke leaves by developing thermoprocessed PLA-based packaging films incorporating ALE obtained through s-SWE. Our hypothesis is that ALE can contribute to improve the bioactivity of PLA-based materials without addition of a downstream process for the extract purification, thus promoting the circularity of artichoke leaves. The influence of ALE on the microstructural and physicochemical properties of PLA films was thoroughly investigated. In addition, the bioactivity of the resulting films was evaluated to assess their potential as sustainable active packaging alternative.

2. Materials and methods

2.1. Materials

Artichoke (*Cynara scolymus*) leaves were provided by Proexport (Murcia, ES) and stored at −20ºC until the usage moment. Chopped leaves with the stems were dried in an oven with convection forced air at 60°C for 48 h, followed by griding, prior extraction. Neat PLA and triethyl citrate were purchased from 123-print (Jordbro, SE) and Sigma Aldrich (Stockholm, SE), respectively.

2.2. Preparation of artichoke leaves extracts by s-SWE

Sequential subcritical water extraction (s-SWE) was applied to biomass powders using an accelerated solvent extractor Dionex ASE 350 (Thermo Fischer Scientific Inc., USA). The extraction cell was prepared with cellulose filter, followed by 3 g of biomass and another layer of cellulose filter. The extraction was carried out using tap water (pH 6.35 ± 0.33), constant pressure (1500 psi) and solid:liquid ratio of 1:11. Herein, fractionation was performed isothermally at 150°C under two sequential cycles of 20 min for optimization of bioactive extract in accordance with Pereira & Jiménez-Quero, 2025. For the extraction procedure, stainless steel cells of 34 mL were used, and the extraction was performed under static mode at fixed volume. ALE fractions were coded as ALE150–20' and ALE150–40', respectively. Fractions from s-SWE were dried in freeze-dryer (FreeZone 6, Labcombo, USA) before characterization.

2.3. Extract characterization

2.3.1. Monosaccharides content

Monosaccharides content in ALE was determined in terms of neutral sugars and uronic acids using a two-steps methanolysis followed by trifluoracetic acid (TFA) hydrolysis in accordance with Willför et al. (2009). Determinations were performed in a High-Performance Anion-Exchange Chromatography coupled to Pulsed Amperometric Detection (HPAEC-PAD) using a Dionex ICS-6000 system (Thermo Fischer Sci., Waltham, USA). A column CarboPac™ PA20 (3 ×150 mm, Thermo Fischer Sci., Waltham, USA) was applied for separation at 0.4 mL/min, using a gradient described in Massironi et al. (2024). Quantification was carried out against calibration curve built with neutral sugars (fucose, arabinose, rhamnose, galactose, glucose, xylose, and mannose) along with uronic acids (galacturonic and glucuronic acids) prepared at different concentrations.

2.3.2. Starch content

Measurement of starch content in the ALE was carried out using the Megazyme enzymatic Total starch kit (Wicklow, IE) and quantified against a glucose standard curve at 510 nm.

2.3.3. Protein content by Bradford

Protein content of ALE was determined using the Bradford method (Bradford, 1976) using a Bio-Rad (Hercules, USA) protein assay kit. Samples were quantified against a calibration curve using Bovine Serum Albumin (Sigma-Aldrich, SE) and the measurement were carried out at 595 nm.

2.3.4. Total phenolic compounds (TPC)

TPC of ALE was determined using Folin Ciocalteu's reagent according to Cicco et al. (2009). Briefly, 25 μL of extracts and 25 μL of Folin-Ciocalteu reagent were added to 96-well microplate. After 2 min, 200 μL of sodium carbonate (5 %; w:v) was added. Microplates were incubated at 37 ºC in the dark for 20 min and read at 760 nm in a FLUOstar Omega (BMG Labtech, DE microplate reader, country). Gallic acid was used as standard, and the results were expressed in mg GAE/g of dry sample.

2.3.5. Phenolic compounds profile

For understanding the phenolic profile present in ALE, samples (10 mg) were submitted to saponification with 500 μL of a 2 M sodium hydroxide solution in an inert atmosphere at 30 °C overnight, followed by adjustment of pH to 2 with HCl. A liquid:liquid partition with ethyl acetate at the proportion of 1:2 (sample:ethyl acetate, v:v) performed in triplicate. Samples were left to evaporate under nitrogen steam until dryness and resuspended in 0.1 % formic acid and methanol mixture (50:50; v:v). Identification and quantification were performed in a HPLC system, Vanquish™ Core (Thermo Fischer Sci., Waltham, USA) coupled to a diode array detector (Thermo Fischer Sci., Waltham, USA). For separation, 25 μL was injected into a reverse phase column Luna 3 μm C18(2) (150 ×4.6 mm; Phenomenex, Torrence, USA) at a flow rate of 0.7 mL/min was used and a gradient separation using 0.1 % formic acid as eluent A and methanol as eluent B applied as follow: 95 % eluent A for 50 min; 50 % eluent A for 15 min and equilibration with 95 % eluent B for 10 min. Quantification was performed against hydroxybenzoic acids (gallic acid, gentisic acid, and vanillic acid) hydroxycinnamic acids (chlorogenic acid, caffeic acid, ρ-coumaric acid, ferulic acid, sinapic acid, and cinnamic acid), and flavonoids (catechin, rutin, quercetin, and luteolin) at 280 nm.

2.4. Thermoprocessing of PLA-ALE materials

Neat PLA was dried at 40°C for 24 h before extrusion, to avoid PLA degradation during processing. PLA, ALE150–40', and triethyl citrate as plasticizer, were blended in accordance with the Table 1. The different materials were processed using a twin-screw extruder (Xplore MC5 micro compounder, Xplore Instruments BV, Sittard, NL) at 145°C and screw speed of 50 rpm in open state. The compounds were mixed just before extrusion and immediately fed into the extruder. All the extrudates samples were applied to compression mold at 145°C with an applied force of 150 kN for 10 min using an automatized compression molding machine (Frontjine presses BV, Rotterdam, NL).

Table 1
Composition of the PLA/artichoke extract (ALE) blends.

Treatment	PLA (%)	Plasticizer (%)	ALE (%)
SP-ALE0	90	10	0
SP-ALE1	89	10	1
SP-ALE2.5	87.5	10	2.5
SP-ALE5	85	10	5

2.5. Films characterization

2.5.1. Scanning Electron Microscopy (SEM)

The morphology of the cryogenically fractured cross-section of the PLA-based films was observed by SEM using a microscopy an Ultra 55 microscope (Zeiss, Oberkochen, DE) in high vacuum SE 2 mode with an accelerating voltage of 10 kV. Samples were sputtered with a 10 nm gold layer.

2.5.2. Color and UV-blocking properties

The color of the film samples was measured using a CR-400 colorimeter (Konica Minolta Chroma Co., Osaka, JPN) and the results expressed in HunterLab color measurement system. For each group, color values were recorded at six random points, and the L*, a*, and b* parameters were obtained. The overall color difference (ΔE) between the control film and the ALE-incorporated samples was calculated using the following equation:

$$\Delta E = \sqrt{\left(L_2^* - L_1^*\right)^2 + \left(a_2^* - a_1^*\right)^2 + \left(b_2^* - b_1^*\right)^2} \quad (1)$$

Where, L_1*, a_1*, and b_1* are the color parameters of the film samples containing ALE, while L_2*, a_2*, and b_2* are the color parameters of the control film.

The UV blocking properties of composite films was investigated by measuring the absorbance of the films using a UV-Vis spectrophotometer (SPECTROstar nano, BMG Labtech, DE), within the wavelength of 250–800 nm and the ultraviolet protection factor (UPF) was assessed by AS/NZS 4399 standard (AS/NZS, 4399, 2017).

$$\mathrm{UPF} = \frac{\sum E(\lambda).S(\lambda).\Delta\lambda}{\sum E(\lambda).S(\lambda).T(\lambda).\Delta\lambda} \quad (2)$$

where E(λ) is relative erythemal spectral effectiveness, S(λ) is solar spectral irradiance, T(λ) is spectral transmittance of the fabric, λ is wavelength (from 280 to 400 nm), and Δλ is wavelength interval. All the color and UV-blocking assays were performed in triplicates.

2.5.3. Fourier transformed infrared spectroscopy (FTIR)

FTIR was performed to identify interactions between the PLA-based materials components. For this, the measurements were carried out from 4000 cm^{-1} to 600 cm^{-1}, using a Spectrum 3 spectrometer (Perkin Elmer, Waltham, USA), equipped with a Universal Attenuated Total Reflectance (ATR) cell device (GladiATR, Pike Technologies, Madison, USA) constituted of a diamond crystal. The measurements were performed by averaging 32 scans with a resolution of 4 cm^{-1}.

2.5.4. Water vapor permeability (WVP)

WVP of the PLA-based films was measured according to the Standard Test Method for Water Vapor Transmission of Materials (ASTM, 2024) at 20°C ± 2°C. The films were sealed at a circular opening of a permeation cell (TQC Sheen, Zuid-Holland, NL), which contained distilled water (RH = 100 %) and was placed in a desiccator filled with silica (RH = 0). The cell was periodically weighted and WVP was determined according to Eq. (3).

$$WVP = \left(\frac{g}{tA}\right)\left(\frac{x}{\Delta P}\right) \quad (3)$$

where A is the permeation area (m^2), g/t is the ratio between mass change (g) and exposition period (s), x is the thickness of the film (m), and ΔP is the difference between water vapor pressure and the vapor pressure of the silica environment (Pa). The assays were performed in triplicates.

2.5.5. Contact angle measurement

Surface hydrophobicity of the films was measured using an Attention Theta Optical Tensiometer (Biolin Scientific AB, SE) under ambient conditions. Deionized water (13 μL) was dropped by a micro syringe onto the film surface. The angle between the drop boundary and film surface was measured using OneAttension software (Biolin Scientific AB, SE) when water released onto the film surface and after 60 s and images of water droplets were saved. Five measurements on the films were conducted to determine the mean value of water contact angle (WCA). The assays were performed in triplicates.

2.5.6. Mechanical properties

Mechanical properties of PLA-based films were investigated in function of the Young Modulus (YM), Tensile strength (TS) and Elongation at the break (EB). The measurements were performed with samples with a shape 5 A form the ISO 527–1 (International Organization for Standardization, 2012) standard, using a Instron 5565 Universal Testing Machine (Instron, Massachusetts, USA) equipped with a load cell of 5 kN. The experiments performed at 20 mm/min. Samples thickness was measured using a digital micrometer (Mitutoyo, Kanagawa, JPN) averaging 5 random points and the assays were performed in triplicates. The assays were performed in quintuplicates.

2.5.7. Thermal properties

Thermal stability of the films was evaluated by thermogravimetric analysis (TGA). The assays were performed in a thermogravimetric analyzer TGA/DSC 3 + Start System (Mettler Toledo, CH) using approximately 10 mg of samples applying a temperature ramp from 25 to 500 °C under a N_2 atmosphere and a heating rate of 10 °C/min.

Differential scanning calorimetry (DSC) was performed for investigation of phase transition using a DSC 2 STARe system (Mettler Toledo, CH). The samples were heated from −20 to 180°C under a N_2 atmosphere with a purge gas rate of 50 mL/min following the program: equilibration at −20°C for 5 min, heating to 180°C at a rate of 20 °C/min; equilibration at 180 °C for 5 min; cooling to −20°C at 20 °C/min, then heating to 180°C at 20°C. Results were evaluated using the software STARe Excellence V13.00 (Mettler Toledo, CH).

2.5.8. Antioxidant activity

The antioxidant activity was measured in terms of the scavenging activity against the radical DPPH (1,1-diphenyl-2-picrylhydrazyl) following the method described by Brand-Williams et al. (1995) with few modifications. The assay was performed with the different PLA-based materials over three cycles of oxidation to determine the antioxidant property performance could be maintained over time. For this, 2 mL of 0.1 mM methanolic solution of DPPH were added to samples (~50 mg), and the results were monitored by three DPPH addition cycles in intervals of 15 min. Samples radical scavenging activity (RSA) were compared with antioxidant standards including gallic acid, ascorbic acid, and ferulic acid submitted to the same conditions of oxidative cycles. The results were determined at 517 nm, using a microplate reader FLUOstar (BMG Labtech, DE).

2.5.9. Antimicrobial activity

The antimicrobial activity of the PLA-based blends was evaluated against common foodborne illness pathogens: *Escherichia coli* (CCUG 10979), and *Staphylococcus aureus* (CCUG 10778) as Gram-positive and Gram-negative representatives, respectively, according to the dynamic contact conditions described by ASTM E2149–20 (ASTM, 2020) with some modifications. The different bacteria inoculum was prepared using lysogeny broth (LB) and incubated at 37°C for 24 h. Optical density at 600 nm of the inoculum were adjusted to McFarland 0.5 standard (~10^8 CFU/mL). Samples (1 ×2 cm^2) were added in tubes containing 5 mL of approximately 10^5 CFU/mL culture and incubated at 37°C for 24 h. An aliquot of 100 μL of the cultures were poured on the surface of solidified Mueller Hinton Agar plates for the viable cell count. The number of colonies was counted after 18 h of incubation at 37°C in comparison with the untreated test specimens (blank).

2.5.10. Overall migration

The overall migration of the film samples was evaluated according to the EU Standard (Regulation (EU) No 10/2011). For this, film specimens (3×3 cm^2) were immersed in 30 mL of food simulants (10 % and 95 % ethanol) representing the aqueous and fatty foods in sealed tubes. The samples were incubated at 40°C for 10 days to simulate long-term storage conditions. After the exposure period, the simulants were evaporated, and the non-volatile residue was weighed gravimetrically. The overall migration was calculated according to Eq. 4 and the values were expressed in mg.dm^{-2}.

$$OM = \frac{(m_a - m_b)}{A} \text{x}100 \quad (4)$$

Where OM is the overall migration (mg/dm^2), m_a is the initial mass of sample after oven drying at 60°C overnight (mg), m_b is the mass of residual film after evaporation of simulant and oven drying at 60°C overnight (mg), A is the area of the test specimen intended to enter in contact with the foodstuff (dm^2).

2.6. Statistical analysis

Statistical significance was calculated using *t*-test or one-way Analysis of Variance (ANOVA) with a significant level of 5 % was applied when appropriate. When ANOVA results were significant, a Fisher's LSD post-hoc test was used to compare group means. The statistical analysis was carried out using R Statistical Software (v4.5.0; R Core Team 2025).

3. Results and discussion

3.1. Physicochemical characterization artichoke leaves extract (ALE)

Sequential subcritical water extraction (s-SWE) was applied as valorization approach for artichoke leaves, since it can facilitate the fractionation of the biomass for extracting bioactive compounds while tailoring the profile of compounds of interest. Physicochemical properties of obtained ALE fractions are shown in Table 2. Carbohydrate content increased by ~7.06 % in the ALE150–40' fraction, mainly characterized by a reduction on glucose levels and increased concentration of galacturonic acid (Fig. S1). Despite the carbohydrate content in both fractions were not statically different ($p > 0.05$), the prominent profile change was consistent with decrease in starch, its polymeric source. This trend is attributed to the high solubility of starch, which is preferentially extracted over other polysaccharides. In the first fractionation step, 266.47 ± 33.57 mg/g of starch was recovered, while more recalcitrant carbohydrates were obtained afterwards. Similar starch removal during fractionation of bay tree pruning waste using s-SWE for extraction of bioactive pectin carbohydrates has been reported (Rincón et al., 2021).

Table 2
Composition of artichoke leaf extract fractions obtained by sequential subcritical water extraction at 150°C for 20 and 40 min.

Compounds	ALE150-20'	ALE150-40'
Yield (mg/g DW)	52.03 ± 1.56^a	4.55 ± 0.10^b
Carbohydrate content (mg/g DW)	404.18 ± 44.08^a	432.85 ± 98.13^a
Starch (mg/g DW)*	266.47 ± 33.57^a	79.27 ± 20.60^b
Total phenolic content (mg/g DW)	24.35 ± 1.96^b	51.07 ± 3.24^a
Soluble protein (mg/g DW)	18.40 ± 0.34^b	43.46 ± 1.39^a
EC_{50} (mg extract DW/mg DPPH)	7.69 ± 0.52^a	3.96 ± 0.79^b

Results are expressed as average (number of replicates = 3) ± standard deviation.
DW refers to dry weigh.
*Starch content determined by enzymatic assay and expressed as part of total carbohydrate content.
Results were statistically compared by t-Test, and the differences were indicated by different letters in the row.

Moreover, a significant increase ($p < 0.05$) in TPC by ~109 % was observed in ALE150–40' compared with ALE150–20', accompanied by a significant decrease ($p < 0.05$) in the EC_{50} value from 7.69 ± 0.52–3.96 ± 0.79 mg extract/mg DPPH, indicating enhanced antioxidant capacity in second fraction. This trend may be attributed to the removal of molecules that hinders the accessibility to the phenolic compounds, such as non-structural carbohydrates, and the improvement of the extraction of phenolic compounds bound to the carbohydrates in the biomass (Cocero et al., 2018). Likewise, Martinez-Fernandez et al., (2021) reported a prominent enhancement of extracts bioactivity after fractionation under s-SWE. Herein, a caffeoylquinic acid-derivative quantified as caffeic acid was the major phenolic compound present in the ALE in both fractions, followed by a slight increase on luteolin content for the second fraction as shown in Fig. S2, agreeing with results previously shown by Mulinacci et al. (2004) for artichoke leaves. Based on the highest amount of TPC and improved bioactivity, ALE150–40' was selected for incorporation on PLA-based active packaging.

3.2. PLA-based active films

3.2.1. Morphology, optical and structural properties

PLA films incorporated with ALE were investigated as alternative materials for packaging materials. For this, it is fundamental to evaluate the properties of the developed materials and the impact of ALE incorporation on the film's properties. The microstructure of PLA-based films cross-sections is shown in Fig. 1a. The presence of ALE in the material can be observed as small inclusions (less than 1 μm), the number of which increases with extract content, as well as larger agglomerates (single μm in size, marked with circles), suggesting a not completely dispersion of ALE during extrusion processing and may contribute to the less compact polymeric matrix. In the case of the plasticizer, there is also no dispersion at the molecular level, which is manifested by the presence of elongated structures resembling fibers on the material's surface.

The optical properties and visual appearance of PLA films changed notably with ALE addition (Fig. 2a). The neat PLA film appeared highly transparent, while films containing ALE exhibited a progressively more brownish tone, indicating the impact of ALE's natural color on the film matrix. This visual trend is supported by quantitative color analysis, summarized in Table S1. With increasing ALE content, the L* values decreased from 85.50 (SP-ALE0) to 65.55 (SP-ALE5), indicating a significant ($p < 0.05$) darkening of the films. Meanwhile, the b* values rising from 0.79 to 28.66, reflects an intensified yellowish-brown coloration in function of ALE concentrations. The total color difference (ΔE) rose from 6.58 to 35.12 with higher ALE concentrations, confirming visible changes in film color.

Evaluating visible light transmittance provides insight into packaging film transparency, while UV-blocking efficiency is crucial for maintaining food quality, as UV radiation accelerates degradation of photo-sensitive compounds. As shown in Fig. 2a, neat PLA exhibited the highest visible transmittance (400–700 nm), whereas incorporating 1, 2.5, and 5 wt% ALE reduced transmittance at 660 nm by 12.50, 60.30, and 71.11 %, respectively (Table S1). Conversely, ALE markedly enhanced UV-blocking across UVA (320–400 nm), UVB (280–320 nm), and UVC (100–280 nm), attributed to its phenolic compounds with UV-absorbing sp^2 and sp^3 benzene ring structures (Gaikwad et al., 2019). This effect is consistent previous report of other phenolic-rich extract (Danmatam et al., 2023). Based on UPF values, films with 5 wt% ALE reached above 20 %, classified as "good" protection (blocking 93.3–95.8 % UV), demonstrating ALE's effectiveness in enhancing UV resistance. Although ALE reduces transparency, its functional properties make it promising for light-sensitive or UV-protective packaging.

All PLA-based films presented characteristics absorption bands around 1750 cm^{-1} (-C═O streching band), 1454 cm^{-1} (-CH_3 bending), 1270 cm^{-1} (-C═O bending), 1183 cm^{-1} (-C-O-C streching), 1045 cm^{-1} (-OH bending), besides the presence of –C–C– stretching bands at 863 cm^{-1} is attributed to the amorphous phase of PLA (Garlotta, 2001;

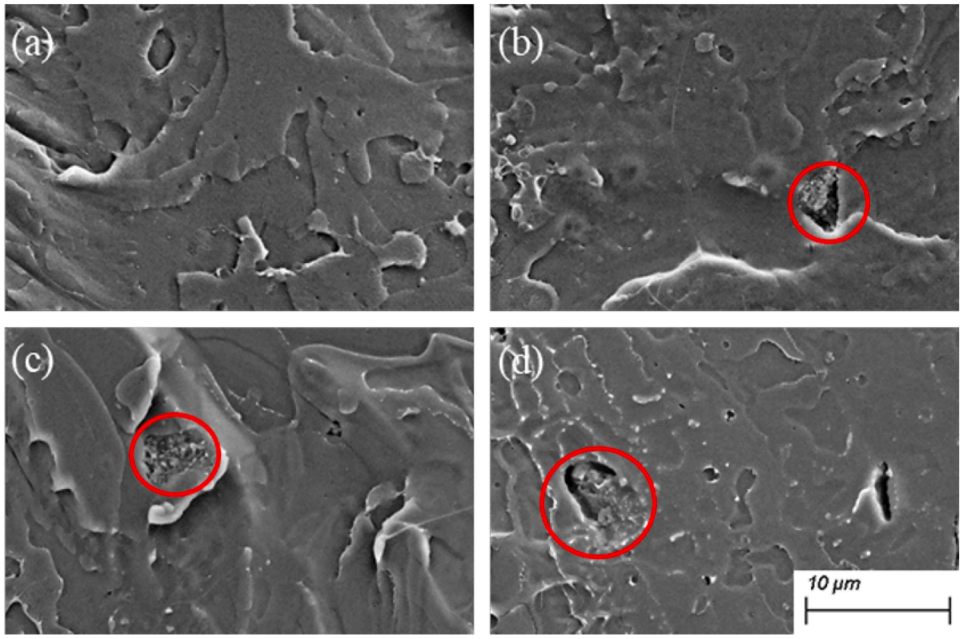

Fig. 1. Cryogenically fractured cross-section micrographs from PLA-based films: **(a)** SP-ALE0, **(b)** SP-ALE1, **(c)** SP-ALE2.5, and **(d)** SP-ALE5.

Martins et al., 2018) as shown in Fig. 2b. With incorporation of ALE, no significant changes were observed in the FTIR sprectra of the PLA-based films. Characteristic band of -OH streching around 3350 cm^{-1} attributed to the high presence of hydroxyls from polyphenols was not evidenced in the FTIR of PLA-ALE films. A similar result was reported by Martins et al., (2018). Likewise, Yaman et al. (2024) reported that the multi-component PLA-based films exhibited no additional characteristic peaks beyond those typically associated with pure PLA. These results suggest that due to the lower amount of extracts on the blend, hydroxyls groups are overlapped by the intensity of the other molecular bonds (Yaman et al., 2024). Besides, a weakening in the band intentisity proportional to the concentration of ALE migh indicate that the extracts contribute to reduce the intramolecular interactions on the polymeric matrix.

3.2.2. *Physicochemical properties*

Water vapor permeability (WVP) is an important barrier property for packaging materials since it has a direct influence on the maintenance of the quality and shelf life of the packaged foodstuffs. The WVP of the PLA-based films ranged from 7.30 to 8.36×10^{-2} g.mm/h.m^2.kPa (Table 3). Similar values of WVP were found by Freitas et al. (2023) for neat PLA films. No significant changes ($p > 0.05$) were observed with the incorporation of ALE on PLA films. These results agreed with the FTIR data, where despite the incorporation of ALE, no prominent available hydroxyls groups were found, which in turn do impact on the water vapor interaction with the polymeric matrix and resulting diffusion.

The water contact angle (WCA) indicates surface wettability of materials, closely aligned to the water adsorption capacity, where a high contact angle corresponds to lower surface wettability. Table 3 shows average WCA measured immediately and 60 s after a water droplet was placed on the surface. For all films, WCA values decreased slightly (2–3°), though not significantly ($p < 0.05$). Neat PLA showed ~74°, consistent with solvent-cast films (Yaman et al., 2024) but lower than extrusion-prepared PLA (>100°) (Hu et al., 2025), likely due to plasticizer migration increasing surface hydrophilicity. Incorporating 2.5 wt % ALE significantly reduced ($p < 0.05$) the angle to 62.41°, which may be attributed to polysaccharides and phenolics orienting at the surface and promoting hydrogen bonding, with possible effects from increased roughness. At 5 wt% ALE, the contact angle rose again, likely due to surface aggregation altering chemistry and morphology, as supported by FTIR and SEM results.

Mechanical properties are critical for packaging performance. Table 3 shows tensile strength (TS), elongation at the break (EB), and Young modulus (YM) of PLA-based films. TS increased significantly ($p < 0.05$) at low ALE content but decreased at higher concentrations, remaining overall comparable to neat PLA. In contrast, all ALE-containing films showed significantly lower EB ($p < 0.05$), with no differences among them, while YM was unaffected ($p > 0.05$). The decrease in EB for samples containing ALE is probably due to the non-ideal dispersion of the extract in PLA and the presence of agglomerates observed in SEM or insufficient adhesion of the ALE particles to PLA and consequently poor stress transfer across the interphase (Piekarska et al., 2016). Similar EB decreases were reported for PLA with phenolic compounds derived from green tea and rosemary (Andrade et al., 2023).

Table 3
Thickness, water vapor permeability (WVP), contact angle (WCA) and mechanical properties of PLA films incorporated with artichoke leaves extract (ALE).

Films	Thickness (mm)	WVP (x10^2 g.mm/h.m^2.kPa)	WCA 0 s (°)	WCA 60 s (°)	Tensile strength (MPa)	Elongation at the break (%)	Young Modulus (MPa)
SP-ALE0	0.19 ± 0.02	8.31 ± 0.46	73.86 ± 2.03^{a}	70.79 ± 1.53^{ab}	46.5 ± 5.0^{ab}	8.2 ± 1.8^{a}	1173 ± 176
SP-ALE1	0.18 ± 0.01	7.77 ± 0.10	68.38 ± 3.81^{b}	67.28 ± 3.81^{b}	51.9 ± 0.4^{a}	5.2 ± 0.6^{b}	1197 ± 52
SP-ALE2.5	0.18 ± 0.02	7.30 ± 1.07	62.41 ± 2.49^{c}	61.11 ± 2.53^{c}	41.1 ± 2.1^{b}	5.3 ± 1.3^{b}	1142 ± 68
SP-ALE5	0.21 ± 0.02	8.36 ± 1.08	70.18 ± 1.89^{ab}	68.89 ± 1.85^{b}	45.5 ± 3.2^{b}	4.8 ± 1.2^{b}	1320 ± 126

Results were expressed as average ± standard deviation.
Letters in the different columns indicate statistical difference among the results.
Results not followed by letters, indicate no statistical significative ANOVA test.

The thermal stability of the materials is shown in Fig. 2c. Neat PLA film exhibited a single degradation step at 337.6°C (T_{max} = 364°C). Compared with neat PLA, PLA-ALE films showed decreasing thermal stability with increasing ALE content: T_{onset} at 334°C for SP-ALE1 (T_{max} = 359°C), 322°C for SP-ALE2.5 (T_{max} = 344°C), and 314°C for SP-ALE5 (T_{max} = 330°C). This indicates reduced stability and a shift toward two-step degradation: 180–250°C (extract degradation) and 300–400°C (PLA chain degradation).

Indeed, the presence of ALE contributed to weakening the intermolecular interactions on PLA, as observed in the FTIR, seems to favor the susceptibility of the materials to the thermal degradation. The same behavior was observed by Acquavia et al. (2024) for PLA-Tea waste blends and by Martin-Perez et al. (2025) for PLA-almond extracts developed materials, where the incorporation of the extracts decreased the thermal stability of PLA films. However, considering most of the food application between 25 and 200°C, the ALE incorporation seems not affect the material thermal integrity.

The thermal behavior of PLA-ALE films assessed by DSC is shown in Fig. 2c. The glass transition temperature (T_g), obtained from the second heating, decreased slightly from 49 to 46°C with increasing ALE, suggesting the extracts increased chain mobility but not enough to act as plasticizers. The thermograms also revealed semicrystalline blends due to an exothermic cold crystallization event (T_{cc}) at 111–113°C. Samples did not crystallize during cooling, and reheating induced cold crystallization, with enthalpy values similar to fusion enthalpy, indicating amorphous samples. The T_{cc} values were similar across samples (Table S2), showing no nucleating effect of ALE. The melting temperature (T_m) ranged from 144 to 148°C, though peak determination may be affected by overlapping melting peaks (α′ and α forms) and possible recrystallization (Gracia-Fernández et al., 2012)

3.2.3. Bioactive properties

Bioactivity in packaging materials is a crucial tool to aid in food preservation and maintenance of quality during storage due to the potential to extend the shelf life of products by preventing accelerated oxidative reactions and microbial growth. The bioactivity of the PLA-ALE films was investigated in terms of DPPH radical scavenging activity, and the results are shown on Fig. 3a.

Neat PLA films showed poor RSA (~9 %), which was statistically similar ($p < 0.05$) to the films with the lowest ALE content. This result indicates that at low levels, ALE does not significantly affect RSA, which is mainly governed by the plasticizer. Ren et al. (2022) reported similar results using acetyl tributyl citrate. In contrast, ALE incorporation at 2.5 and 5 % produced high RSA (41.56–29.76 % for SP-ALE2.5 and 92.67–70.60 % for SP-ALE5) across three oxidative cycles, comparable to the common antioxidants: gallic, ascorbic, and ferulic acids. SP-ALE5 initially matched gallic acid ($p < 0.05$), confirming strong activity. For this sample, the RSA decreased with cycles, stabilizing at 75.51–70.60 % from the second oxidative cycles with RSA values consistent with previous report using natural extract in PLA (Acquavia et al., 2024). This effect might be related to the high phenolic content of ALE (Table 2), which tend to slow release from the PLA matrix until reaches stability.

Complementarily, the antimicrobial activity of the films was evaluated against Gram-negative and Gram-positive bacteria (Fig. 3b). PLA-

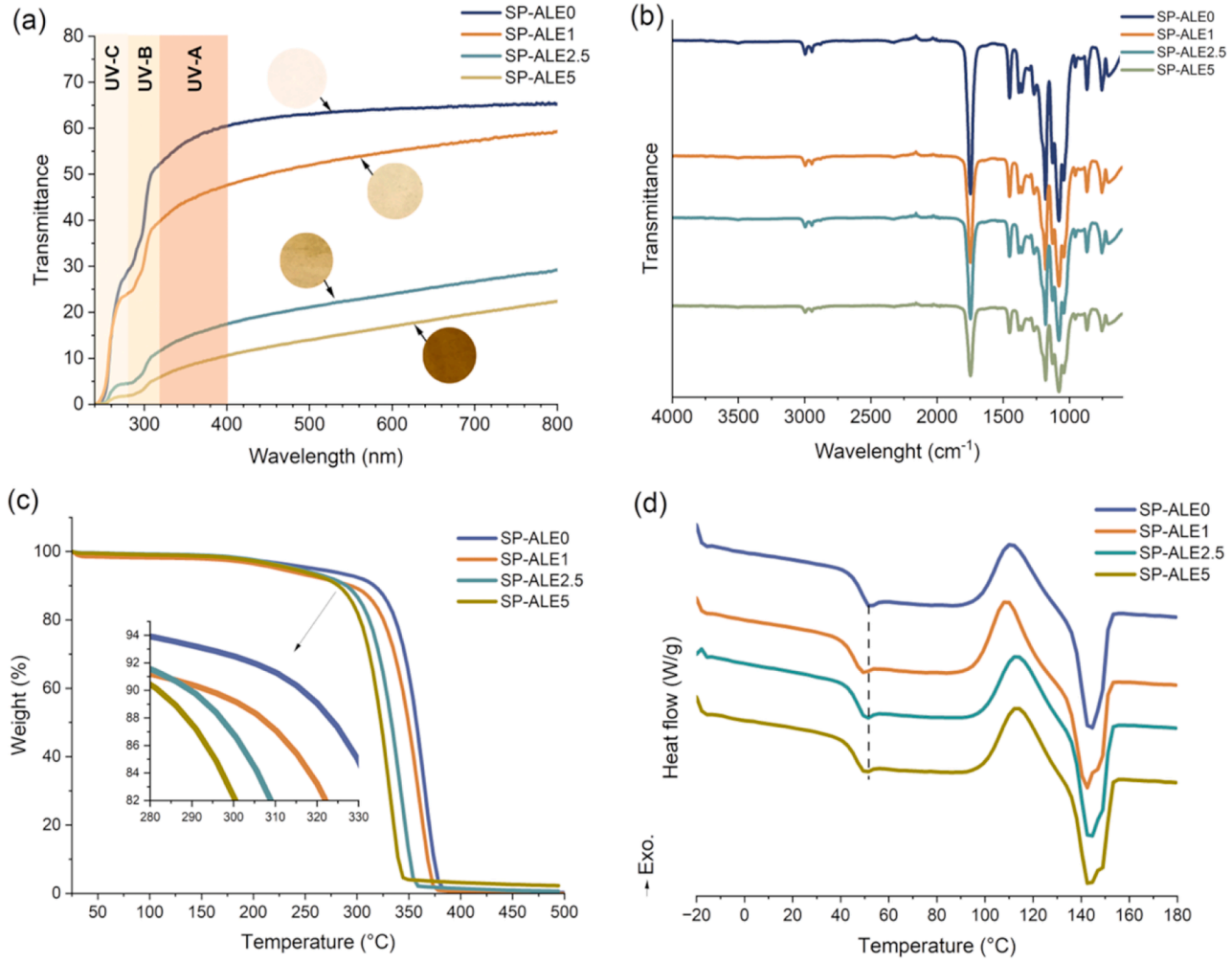

Fig. 2. **(a)** UV–vis spectra; **(b)** FTIR spectra; (c) TGA, and (d) DSC thermograms of the different PLA-based films incorporated with artichoke leaves extract.

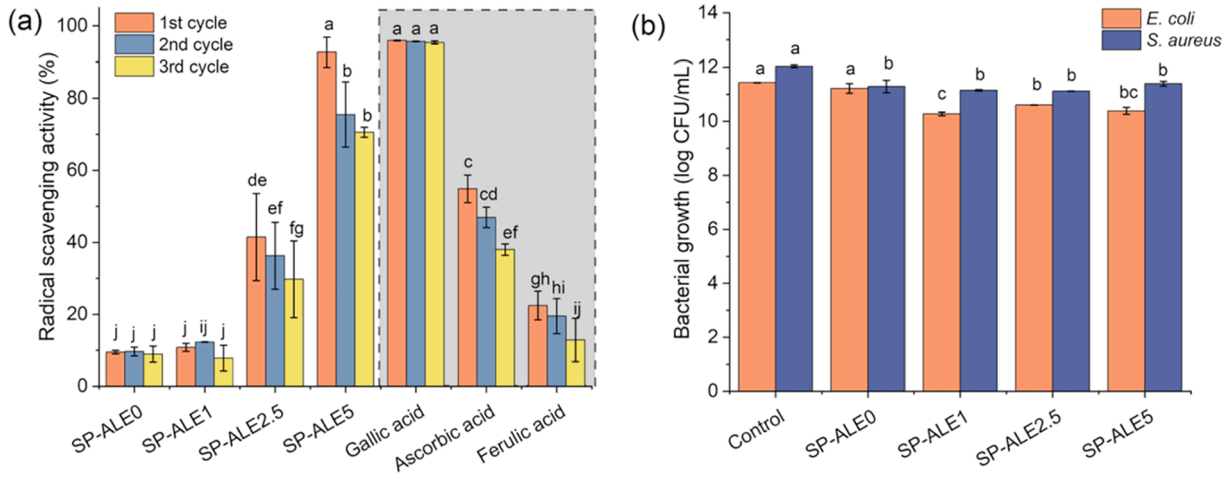

Fig. 3. **(a)** Radical scavenging activity of the different PLA-based films containing ALE in comparison with common antioxidant agents (gallic, ascorbic and ferulic acids), and **(b)** antimicrobial activity of the materials against *E. coli* and *S. aureus*.

ALE films showed growth inhibition of *E. coli* and *S. aureus*, with differences reflecting their cell wall structures: Gram-negative bacteria such as *E. coli* have a peptidoglycan layer plus an outer lipopolysaccharide membrane, whereas Gram-positive bacteria such as *S. aureus* are generally more susceptible to antimicrobial agents in comparison with Gram-negative bacteria (Swebocki et al., 2024). PLA-based films presented 0.21–1.15 log reduction against *E. coli* and 0.65–0.93 log reduction against *S. aureus,* depending on ALE concentration. For *E. coli*, addition of extracts of 1 % to PLA presented the highest inhibition, not statistically distinct ($p < 0.05$) from ALE incorporation at 5 %. Similar log reduction was previously reported by Ordonez et al. (2022). Whereas for *S. aureus*, notably, SP-ALE0 showed 0.75 log reduction, suggesting that triethyl citrate plasticizer contributes to antimicrobial activity of the Gram-positive bacteria instead ALE, in agreement with Karabagias et al. (2024) who reported similar effects with tetraethyl citrate.

3.2.4. Migration evaluation

The overall migration of PLA-ALE films was evaluated in food simulants (10 % and 95 % ethanol) as shown in Fig. 4. Migration was significantly affected by both simulant type and ALE concentration. The interaction between the polymer and the simulant, which causes swelling and relaxation of the polymer matrix, plays a key role in determining the extent of compound migration (Martin-Perez et al., 2025). In contact with 10 % ethanol, neat PLA release was minimal, probably related to the release of plasticizer. However, in PLA-ALE films, the partial relaxation of the hydrophobic PLA matrix facilitated the diffusion of non-polymeric components, such as ALE, into the medium in a concentration-dependent manner. Notably, the incorporation of up to 2.5 % ALE resulted in an overall migration below the regulatory limit of 10 mg/dm^2 present at EC No 10/2011 (European Commission, 2011), allowing the PLA-ALE materials to stablish safety and regulatory compliance under this specific application.

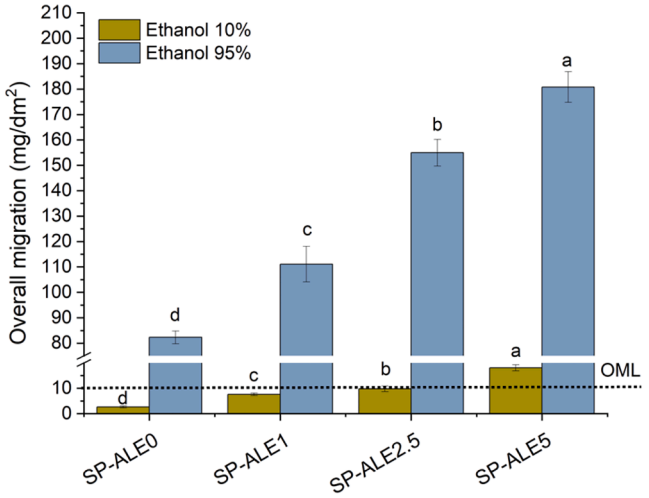

Fig. 4. Overall migration of components from PLA-based films incorporated with different concentrations of ALE films into food simulants (10 % and 95 % ethanol), where OML means overall migration limit.

Although phenolic compounds are not listed in the regulation (European Commission, 2011), they may qualify as food constituents under the regulation No 450/2009 on active packaging (European Commission, 2009). According to this regulation, if an active substance contributes to the functionality of the material, its released amount may be excluded from the overall migration limit. For less polar environment (95 % ethanol), solvent penetration caused swelling and relaxation of the PLA matrix, enhancing diffusion of entrapped molecules. Similar enhanced extraction of phenolics by ethanol has been previously reported (Foong et al., 2025). Interestingly, even neat PLA exhibited high migration in 95 % ethanol, suggesting potential partial degradation of PLA chains upon exposure to ethanolic environments, which may further promote the release of compounds. This phenomenon has also been observed in previous studies (Jamshidian et al., 2012).

4. Conclusion

A novel strategy for valorizing artichoke leaf waste as a bioactive ingredient in functional packaging was proven. Bioactive ALE were obtained through a sustainable s-SWE and incorporated into PLA *via* thermoprocessing. The s-SWE process yielded ALE with reduced starch, high phenolic content, and enhanced antioxidant activity, especially in the second fractionation (ALE 150–40'). This green approach provides functional extracts for bio-based active packaging. PLA-ALE films showed less compact morphology and weaker intermolecular PLA bonds as ALE content increased. ALE addition improved UV protection, while thermal, barrier, and mechanical properties remained largely unaffected. Antioxidant and antimicrobial activities were significantly enhanced, and migration tests confirmed suitability for hydrophilic foodstuffs, with 2.5 % ALE showing low migration into 10 % ethanol, compliant with EU food contact regulations. ALE-based PLA films showed bioactive properties that suggest potential for food preservation applications, particularly without drastically compromising the main properties of PLA. In this way, ALE represents a promising upcycling approach for packaging material development, highlighting a potential

valorization route.

CRediT authorship contribution statement

Pamela F. M. Pereira: Writing – review & editing, Writing – original draft, Visualization, Validation, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Zeinab Qazanfarzadeh:** Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis. **Przemyslaw Sowinski:** Writing – review & editing, Writing – original draft, Visualization, Validation, Formal analysis. **Amparo Jiménez-Quero:** Writing – review & editing, Writing – original draft, Validation, Supervision, Project administration, Funding acquisition, Conceptualization.

Declaration of Generative AI and AI-assisted technologies in the writing process

The authors declare that no AI tool was used in this manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have apeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.fpsl.2025.101686.

Data Availability

Data will be made available on request.

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