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Modulating the properties of bioactive cellulose acetate electrospun films using *aloe vera* gel and lemongrass essential oil for sustainable active food packaging

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Background and aims: Transitioning towards sustainable diets and circular food systems requires bio-based packaging that preserves food quality. This study investigates electrospun bioactive films made from cellulose acetate (CA), *aloe vera* gel (AG), and lemongrass essential oil (EO), with a focus on how variations in composition influence film performance relevant to active food packaging.

Methods: Three formulations with distinct mechanical profiles (rigid, flexible, and balanced) were fabricated by electrospinning solutions of CA, AG and EO. Morphology (visual appearance, SEM), molecular interactions and structural order (FTIR, XRD), thermal behavior (DSC, TGA), barrier properties (moisture loss, solubility, swelling index, water vapor permeability), optical performance (color, light transmission, UV-visible shielding), surface wettability (contact angle), and bio-activity (DPPH and ABTS assays; antibacterial tests against representative Gram-positive and Gram-negative strains) were evaluated.

Results: The prepared films showed dense, compact, mat-like morphologies and were mainly amorphous in nature without any chemical degradation of CA. The flexible (AG-rich) films retained more moisture, showed lower thermal transition temperature, higher water solubility, greater swelling-induced mass loss, and higher water vapor permeability. In contrast, rigid and balance films (containing relatively higher CA and EO levels), demonstrated better dimensional stability and resistance to moisture. The optical measurements showed tunable opacity, UV-visible shielding, and wettability, while the EO-enriched balanced film exhibited the highest antioxidant capacity and broad-spectrum antibacterial activity.

Conclusion: Through regulating CA-AG-EO composition, it is possible to manage the mechanical, thermal, barrier, optical, and bioactive properties in electrospun films at the same time. These cellulose acetate-based, multifunctional materials are promising candidates for active food packaging and support circular and more sustainable food systems.

KEYWORDS

active packaging, *aloe vera* gel, cellulose acetate, electrospun films, essential oil

1 Introduction

Growing environmental concerns associated with petroleum-derived plastics have raised global efforts to develop biodegradable and bioactive materials for more sustainable food-packaging applications (Choudhury et al., 2022; Yin and Woo, 2024). In this regard, biopolymers such as starch, chitosan, polylactic acid, and cellulose derivatives have received considerable attention due to their renewability, safety, and tunable physicochemical properties (Dutta and Sit, 2024; Ferreira and Monteiro, 2023). Among these, cellulose acetate (CA) stands out for its excellent film-forming ability, mechanical strength, and compatibility with food-contact systems (Angel et al., 2020). However, CA-based films have limited extensibility with high stiffness, which restricts their use in flexible packaging formats (Park et al., 2024). To address this concern, plasticizers such as glycerol are commonly introduced in CA-based systems to improve flexibility by increasing polymer chain mobility (Catel et al., 2025).

Alongside mechanical optimization, there is increasing attention towards incorporating bioactive compounds into packaging materials to provide antioxidant and antimicrobial properties that are desirable for active food packaging (Jayakrishnan et al., 2024). This has led to the incorporation of naturally derived bioactive compounds, such as *aloe vera* gel (AG), essential oil (EO), or other plant extracts. The combined CA-AG-EO system, can enhance functionality at the food-packaging interface, while also influencing the mechanical, structural, and barrier behavior of the films.

The technique of electrospinning has emerged as a promising to produce polymeric films and fibrous mats of high surface area, tunable morphologies, and potential functionality for food-packaging applications. Electrospinning is an alternative way of making film compared to conventional film-forming techniques, which allows to maintain structural uniformity and functional performance while introducing bioactive compounds in the polymer matrix (Ali et al., 2026). Therefore, electrospun CA-based films can serve as suitable platform to create active packaging materials with specific mechanical and functional properties. To systematically design CA-AG-EO electrospun films with packaging-relevant mechanical performance, a data-driven formulation strategy based on a space-filling Latin Hypercube Design was first adopted. The resulting electrospun films were evaluated for their tensile strength, elongation at break, and Young's modulus. The response surface modelling was applied to quantify the relative contributions and interactions of CA, AG, and EO. CA emerged as the primary structural component responsible for strength and stiffness, in contrast, AG mainly acted as a natural plasticizer that enhanced the film's flexibility while decreasing its modulus. EO showed a composition-dependent effect: at lower levels, it behaved as a mild plasticizer, whereas at higher levels, in specific combinations with CA and AG, it began to disrupt the mechanical integrity of the film. Therefore, based on the mechanical thresholds derived from the experimental data, the films were classified into three representative categories: rigid films with high modulus and low elongation at break, flexible films with high elongation at break and low modulus, and balanced films with intermediate combinations of tensile strength, elongation at break, and modulus. This mechanical mapping served as a practical framework for selecting formulations with mechanical profiles for specific applications.

In the present work, one formulation from each mechanically defined category was selected, and three representative CA-AG-EO films, namely rigid, flexible, and balanced, were fabricated under the same electrospinning conditions. The idea was not just to compare

them but also establish a link between formulation and their behavior. To achieve this, relatively extensive characterizations were conducted, including physical, thermal, structural and functional features. Differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA) were used for thermal characterization. Structural and morphological analyses of the films were made using optical images, scanning electron microscopy (SEM), Fourier transform infrared spectroscopy (FTIR) and X-ray diffraction (XRD). The barrier properties to water, transparency, and the wettability of the films were also evaluated, as well as their antioxidant and antimicrobial properties. This comparison highlights how formulation modulates film performance and demonstrates the potential of CA-AG-EO electrospun films as multifunctional materials for sustainable active food packaging.

2 Materials and methods

2.1 Materials

All the chemical and reagents used in this study were of analytical grade. Cellulose acetate (molecular weight 5,000 g/mol), glycerol. Acetic acid, methanol ($\geq 99.8\%$ purity), for antioxidant analysis: 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) were purchased from Sigma-Aldrich (USA). Lemongrass essential oil was graciously provided by the Central Institute of Medicinal and Aromatic Plants (CSIR-CIMAP), India. The fresh *aloe vera* leaves were collected from Maqam Campus of United Arab Emirates University, Al Ain, UAE. The gel was manually extracted, mixed, and then strained to remove suspended particles. It was then stored at $4 \pm 1^\circ\text{C}$ to preserve its biological activity and for later use. To evaluate the antimicrobial potential of the developed films, two Gram-positive (*Bacillus cereus* ATCC 11778 and *Staphylococcus aureus* ATCC 25923) and two Gram-negative (*Salmonella enterica* ATCC 14028 and *Escherichia coli* ATCC 25922) bacterial strains were selected. All strains were obtained from the American Type Culture Collection (ATCC). For microbiological assays, standard growth media were utilized, including Mueller-Hinton Broth (Hi-Media, Mumbai, India), peptone water, and Plate Count Agar, the latter two were obtained from AcuMedia (Heywood, UK).

2.2 Electrospinning setup and film fabrication

Electrospun CA-AG-EO films were fabricated using an EF300 electrospinning machine (SKE Research Equipment, Italy) under ambient laboratory conditions, at around $22^\circ\text{C} \pm 2^\circ\text{C}$. The prepared spinning solutions were loaded into 50 mL syringes fitted with 20-gauge metallic blunt-end needles. The electrospinning was carried out at a voltage of 25 kV, with a tip-to-collector distance of 12 cm. The flow rate of the solution was maintained at 10 mL h^{-1} , while the collector was rotated at 400 rpm. The spinning process continued for 6 h to obtain uniform, continuous films.

A relatively higher flow rate was intentionally used as preliminary trials have shown that lower flow rates ($1\text{--}3\text{ mL h}^{-1}$) produced highly porous nanofibrous mats. The resulting electrospun CA-AG-EO films were collected on an aluminum foil and stored at $4^\circ\text{C} \pm 1^\circ\text{C}$ for further structural, thermal, physicochemical, optical, antioxidant, and antimicrobial analysis.

Three formulations with distinctive mechanical characteristics were selected for detailed analysis. With a Young's modulus of approximately 75–80 MPa, the rigid film (5.7% CA, 10% AG, 0.3% EO) exhibited strong stiffness but limited extensibility (elongation at break $\approx 12\%$). Alternatively, the flexible film (5.1% CA, 25% AG, 1.2% EO) exhibited markedly higher extensibility (elongation at break $\approx 57\%$) and lower modulus (≈ 15 MPa). The balanced film (4.2% CA, 12.5% AG, 1.5% EO) demonstrated intermediate mechanical behavior, with moderate elongation ($\approx 24\%$) and modulus (≈ 21 MPa). These representative formulations were chosen to allow for comparative analysis of structure–property–function relationships across films with contrastingly different mechanical properties.

2.3 Physical and water barrier characterization

2.3.1 Moisture loss and water solubility

To evaluate the moisture loss, the film samples were cut into 3×2 cm squares, and their initial weights were recorded. These samples were then dried in an oven at 105°C for 48 h, followed by reweighing to determine the final mass. The moisture loss (%) was calculated using Equation 1 as follows:

$$\text{Moisture Loss (\%)} = \left(\frac{W_i - W_f}{W_i} \right) \times 100 \quad (1)$$

where, W_i and W_f are the initial and final weights of a film sample, respectively.

For the determination of water solubility, film samples were cut into 2×2 cm squares and immersed in 30 mL of deionized water at 25°C for 24 h. After incubation, the mixtures were centrifuged at 3000 rpm for 10 min to separate the dissolved components. The supernatant was decanted, and the remaining film residues were dried at 50°C until a constant weight was achieved. The solubility (%) was calculated using Equation 2 as follows:

$$\text{Solubility (\%)} = \left(\frac{W_i - W_f}{W_i} \right) \times 100 \quad (2)$$

where, W_i and W_f are the weights of the original film sample and the remaining film residues after drying, respectively.

2.3.2 Swelling index

The swelling index of the films was assessed by cutting them into 3×2 cm strips and immersing each sample in 50 mL of deionized water for 30 s at room temperature. After soaking, the films were gently blotted to remove surface water and immediately weighed (Fathi Achachlouei and Zahedi, 2018). The swelling index (SI) was calculated using Equation 3:

$$\text{Swelling Index (\%)} = \left(\frac{W_s - W_d}{W_d} \right) \times 100 \quad (3)$$

where, W_s is the swollen weight of the film (after immersion), and W_d is the initial dry weight of the film.

2.3.3 Water vapor permeability

The water vapor permeability (WVP) of the films was assessed following Chen et al. (2020) with slight modifications. Each film sample was tightly sealed over the mouth of a 50 mL Falcon tube holding 40 mL of deionized water, ensuring sufficient air gap of approximately 1 cm between the liquid surface and the inner film surface. The sealed tubes were placed in a desiccator filled with activated silica gel to achieve a controlled low-humidity environment. The whole setup was kept at room temperature, and the tubes were weighed after 24 and 48 h to determine water vapor loss. The WVP, $\text{g}\cdot\text{m}/(\text{m}^2\cdot\text{Pa}\cdot\text{s})$, was calculated using the rate of water vapor transmission through the film as follows:

$$\text{WVP} = \frac{\Delta m \times L}{A \times t \times \Delta P} \quad (4)$$

Where, Δm is the mass of water lost with time, L represents the film thickness, A is the surface area of the film exposed to the vapor, t is the duration in s, and ΔP represents the vapor pressure gradient across the film.

2.3.4 Color attributes and UV-visible optical properties

The color attributes of the films were analyzed using a HunterLab ColorFlex EZ colorimeter, following the CIE Lab color space system. In this system, L^* denotes lightness, a^* indicates the red-green spectrum, and b^* reflects the blue-yellow spectrum. The total color difference (ΔE) was calculated using Equation 5 as follows:

$$\Delta E = \sqrt{(L_t^* - L_0^*)^2 + (a_t^* - a_0^*)^2 + (b_t^* - b_0^*)^2} \quad (5)$$

where, L_0^* , a_0^* , and b_0^* represent the baseline color values of the standard sample (94.29, -1.23 , and 0.21 , respectively), while L_t^* , a_t^* , and b_t^* correspond to the measurements recorded at a specific time point t .

The optical transparency (opacity) of the films was evaluated by recording their transmittance across the wavelength range of 300–800 nm using a UV–Visible microplate spectrophotometer (Multiskan Sky, Thermo Scientific, Sweden), in accordance with the procedure reported by Nagarajan et al. (2012). Film samples were cut into 2×2 cm squares and placed directly into the instrument for color measurement. The overall transparency and UV-blocking capacity were assessed using Equation 6 as follows:

$$\text{Transparency (\%)} = \frac{-\log(T_{600})}{x} \quad (6)$$

where, T_{600} represents the transmittance measured at 600 nm, and x denotes the film thickness (in mm). A lower transparency value indicates greater light-blocking capacity in the CA-AG-EO films.

2.3.5 Contact angle

A custom-built setup was used to measure the water contact angle of the film surfaces. Images of water droplets on the films were

captured using a USB digital microscope with built in LED illumination, operated through Microcapture Plus software (v1.01). A water droplet was gently released from a height of 1 cm through a 22-gauge needle of inner and outer diameter 0.406 mm and 0.711 mm, respectively. This setup was connected to a Fusion 200 syringe pump (Chemyx Inc., Stafford, USA) via Tygon tubing (1/16" internal diameter), and the dispensing rate was maintained at 0.06 mL/min. Further, the contact angle was determined by drawing a tangent to the droplet's edge using ImageJ software (Phasaludeen et al., 2026)

2.4 Structural and morphological characterization of film

2.4.1 Fourier transform infrared spectroscopy

To examine the chemical characteristics of the film samples, Fourier transform infrared (FTIR) spectroscopy (Perkin-Elmer Spectrum, USA) was used. Spectral data were recorded in the range of 450–4,000 cm^{-1} at a resolution of 1 cm^{-1} , with an average of 32 scans per sample. The potential molecular interactions between CA, EO, and AG within the films were assessed by analyzing the resulting absorption bands.

2.4.2 X-ray diffraction

X-ray diffraction (XRD) analysis was performed using an XPRT-3 diffractometer (Malvern Panalytical) to determine how the different material components used affected the crystalline structure of the films. Measurements were performed using Cu-K α radiation ($\lambda = 1.5406 \text{ \AA}$) operated at 40 kV and 30 mA. To ensure high-resolution peak detection, diffraction patterns were recorded in the 2θ range of 10° to 80° with a scanning rate of $0.02^\circ/\text{sec}$.

2.4.3 Environmental scanning electron microscopy

Environmental scanning electron microscopy (ESEM) (Quanta 250 FEG, FEI, Hillsboro, Oregon, USA) was used to examine the films' surface structure. The detail surface structure and topography were observed at an acceleration voltage of 10 kV and a magnification of $5,000\times$.

2.4.4 Differential scanning calorimeter

Differential scanning calorimeter (DSC) Q100 (TA Instruments, USA) was used to study the thermal properties of the films. Each sample (about 5 mg) was hermetically sealed in an aluminum pan. The samples were heated in a nitrogen atmosphere with a flow rate of 50 mL/min. The samples were heated from 40°C to 400°C at a rate of $10^\circ\text{C}/\text{min}$. The instrument was calibrated using a pure indium standard.

2.4.5 Thermogravimetric analysis

To assess the thermal stability of the film samples, they were analyzed by using thermogravimetric analysis (TGA) using a Q50 thermal analyzer (TA 202 Instruments, USA). The samples (about 5 mg) were heated from 25°C to 800°C at $20^\circ\text{C}/\text{min}$ under a constant nitrogen flow. The TGA curves presented the weight loss of film samples as a function of temperature, providing an insight into their thermal degradation pattern.

2.5 Bioactivity assessment

2.5.1 Antioxidant capacity

For sample preparation, 100 mg of films were immersed in 3 mL of (90%) methanol, vortexed for 10 min, followed by centrifugation at 8000 g for 15 min. The film extracts were evaluated using the DPPH and ABTS radical scavenging assays with slight modifications to previously reported method (Rabbani et al., 2025).

Briefly, 0.5 mL of different concentrations of extract was mixed with 0.5 mL of either DPPH or ABTS radical solution, and the mixture was incubated in the dark for 30 min. A 0.3 mL aliquot of the resulting solution was pipetted into a 96-well microplate, and the absorbance was recorded at 517 nm and 734 nm for DPPH and ABTS, respectively. The half-maximal inhibitory concentration (IC_{50}) for the films was calculated and compared with that of gallic acid. Results are expressed as $\mu\text{g GAE/g}$ of film.

For DPPH analysis, 90% methanol, while for ABTS assay, distilled water was used as the solvent. Prior to use, both radical solutions were diluted with their respective solvents to achieve a baseline absorbance near to 1. The DPPH solution (0.004%) was freshly prepared, while the ABTS stock was made by combining equal parts of 7 mM ABTS and 2.5 mM potassium persulfate, which were left to react overnight at $4^\circ\text{C} \pm 2^\circ\text{C}$ in the dark.

2.5.2 Antimicrobial activity

The antibacterial activity of the films was evaluated using the agar disk diffusion method following the method of Ebrahimzadeh et al. (2021). This method was selected because it is suitable for evaluating solid film samples, where the diffusion of antimicrobial compounds from the film into the agar medium is relevant. Briefly, the bacterial suspensions of *Bacillus cereus*, *Staphylococcus aureus*, *Salmonella enterica*, and *Escherichia coli* were prepared and standardized according to 0.5 McFarland turbidity standard which corresponds to about $5 \times 10^6 \text{ CFU/mL}$. The prepared suspensions were evenly spread onto the surface of solidified Mueller-Hinton agar plates with the help of a glass spreader. Under sterile conditions, the sample film discs were cut into 10 mm and gently placed onto the inoculated agar plate surfaces. Commercial antibiotic discs were used as controls. All the films were tested in triplicates. The plates were incubated at 37°C for 24 h, after which the antibacterial activity was determined by measuring the diameter of the inhibition zones (mm) formed around the film discs. The zones of clearance, indicating microbial growth inhibition, were measured using an automated colony counter (Scan 1,200-HD, Interscience, France).

2.6 Statistical analysis

All experiments in this study were conducted in triplicate, and results are reported as mean $\pm$ standard deviation. Statistical analyses were performed using JMP Pro 15 (SAS Institute Inc., USA). One-way analysis of variance (ANOVA) was applied to know significant differences among film formulations for the measured properties. After significant effects were observed ($p < 0.05$), mean comparisons were performed using Tukey's Honestly Significant Difference (HSD) *post hoc* test at a 5% significance level.

3 Results and discussion

3.1 Structural and morphological

3.1.1 Morphological characteristics and appearance

Optical images of the CA-AG-EO films are shown in [Figure 1](#). The rigid film was translucent with a compact and uniform white matrix that allowed noticeable visibility. The high CA content in this formulation is responsible for its dense visual structure and reduced transparency ([Mikaeili and Gouma, 2018](#)). In contrast, flexible film was soft, less dense, and displayed more visibility indicating improved light transmission through the film's matrix, owing to higher fraction of transparent AG ([Cazón and Vázquez, 2021](#); [Sui Chin et al., 2017](#)). Balanced film was opaque with a uniform, smooth, and glossy surface, due to the higher EO content. The presence of EO imparts a faintly yellowish tint and enhances surface smoothness of the film through plasticization ([Ben Dlala et al., 2025](#); [de Oliveira et al., 2020](#)). These differences in opacity, surface uniformity, and light transmission directly reflect the compositional variations in CA, AG, and EO.

In all three films, SEM images showed a mostly mat-like, interconnected structure rather than nanofibers. This structural appearance is due to the relatively high flow rate (10 mL/h) and the short distance from the tip to the collector (12 cm). Under these conditions, the polymer jet undergoes insufficient solvent evaporation during flight, causing the deposited fibers to reach the collector in a partially wet state. As a consequence, fibers readily coalesce, merge, and flatten, forming a continuous mat rather than a porous nanofibrous network. Such fusion is well documented in the literature, where increased flow rate accelerates solution throughput and suppresses jet stretching, both of which promote fiber-fiber merging and dense film formation ([Harriet and Mouriño, 2025](#); [Naeim et al., 2025](#)). While this type of fused structure is different from the highly porous electrospun nanofibrous structures commonly seen at low flow rates, it can be beneficial and intentional within the objectives of this study. The purpose of developing CA-AG-EO films is to create active food packaging. This type of packaging demands a non-porous or low-porosity structure to minimize water vapor transmission, prevent microbial entry, and ensure barrier efficiency ([Lu et al., 2024](#); [Udana Eranda et al., 2025](#)). A porous fiber mat would not possess these critical properties due to increased porosity and lower mechanical strength ([Menkhaus and Fong, 2019](#)).

Hence, the mat-like structure is suitable for the electrospinning process and facilitates the production of dense, cohesive and stable packaging films.

Although all films display a mat-like morphology, the structure of each film also displays distinct characteristics. For example, the rigid film showed a dense, compact and smooth surface, with uniformly distributed small circular pores and dispersed particulate features. The high CA content promotes strong intermolecular interactions and reduces polymer chain mobility, leading to a rigid, cohesive and a stiff network ([del Gaudio et al., 2021](#)). In contrast, flexible films exhibited apparently less dense and open structures, with larger micro-voids and irregular-shaped cavities.

The substantially higher AG content enhances molecular mobility and imparts significant plasticization ([Mujawar et al., 2025](#)). The presence of discrete droplets and dispersed phases further indicates areas where EO may have migrated or become encapsulated within hydrated polymer domains, reinforcing the flexible behavior of the film ([Ben Dlala et al., 2025](#)). Balanced films exhibited irregular, rough morphologies with crystalline or petal-like surface features. These characteristics are associated with its high EO content (1.5%), which might have migrated to the surface or crystallized during drying, resulting in promoting the surface roughness. The surface depressions observed on microcapsules formed during electrospinning are a natural result of polymer shell formation followed by shrinkage during solvent evaporation ([Alehosseini et al., 2018](#)).

3.1.2 Fourier transform infrared spectroscopy

The FTIR spectra of the three mechanically classified films are presented in [Figure 2a](#), over the range of 4,000–500 cm^{-1} . FTIR analysis was employed to confirm the chemical integrity of CA and to assess whether the incorporation of AG and EO introduced detectable changes in the local chemical environment of characteristic functional groups. The strong peak observed at $\sim 1,740 \text{ cm}^{-1}$ corresponds to the ester carbonyl ($\text{C}=\text{O}$) stretching vibration, confirming the integrity of the acetylated cellulose backbone ([Araújo et al., 2020](#)). The prominent bands at $1,235 \text{ cm}^{-1}$ and $1,050 \text{ cm}^{-1}$ are attributed to $\text{C}-\text{O}-\text{C}$ asymmetric stretching and $\text{C}-\text{O}$ stretching vibrations, respectively, which are typical of CA and polysaccharide structures ([Teixeira et al., 2021](#)). The absence of significant peak shifts or changes in band positions in these regions indicates that the chemical structure of CA remains unaffected by electrospinning and additive incorporation, in

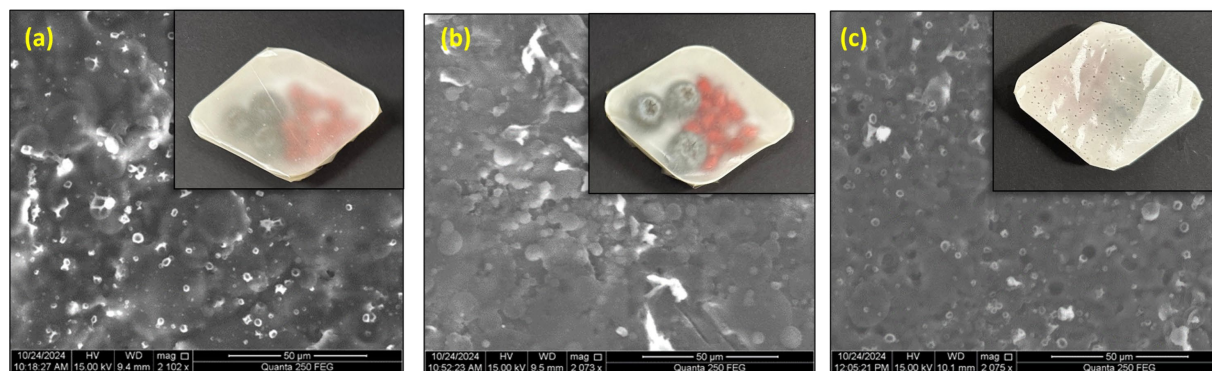


FIGURE 1
SEM images of electrospun CA-AG-EO films: (a) Rigid, (b) flexible, and (c) balanced. Insets show the corresponding visual appearance of each film.

agreement with previous reports on electrospun CA-based (El Fawal et al., 2019).

A broad absorption band in the 3,250–3,550 cm^{-1} region, assigned to O–H stretching vibrations, was observed in all samples, due to residual hydroxyl groups in CA and the abundant hydroxyl groups of AG polysaccharides (Liu et al., 2024; Rahman et al., 2024). Although an increase in band width and intensity was observed with increasing AG content from rigid film to balanced film, and was most pronounced in flexible film, no corresponding or systematic shifts were detected in the C=O or C–O–C bands. From a spectroscopic perspective, this indicates that any hydrogen bonding introduced by AG is weak, highly distributed, or transient in nature and does not significantly perturb the vibrational environment of the ester or ether groups of CA. The C–H bending vibration at 1,430 cm^{-1} was visible in all films and showed a noticeable intensity increase in balanced film, which contains the highest EO content (1.5%). This band is associated with the methyl and methylene groups of terpenoid compounds present in EO, particularly citral and geraniol. The enhanced intensity of this band in the balanced film confirms the successful incorporation of EO into the CA network. This is supported by FTIR spectra reported for essential-oil-loaded biopolymer films (Bittencourt et al., 2025). Importantly, while minor intensity variations were detected across spectra, the lack of discernible peak shifts or new absorption bands, particularly in the C=O and C–O regions demonstrates that FTIR does not provide direct evidence of strong or specific intermolecular hydrogen bonding among CA, AG, and EO. Overall, FTIR analysis confirms that the electrospinning process preserves the

chemical structure of CA and that interactions among CA, AG, and EO are predominantly physical.

3.1.3 Differential scanning calorimetry

DSC was used to investigate the thermal transitions of the CA-AG-EO films. The thermograms revealed characteristic thermal events, including moisture-loss endotherms, the glass transition temperature (T_g), and melting-like or thermal relaxation endotherms (T_m), as shown in Figure 2b. All three samples showed an initial broad endothermic region below 100 $^{\circ}\text{C}$, which is attributed to the loss of physical and hydrogen-bonded water present within the films. This low-temperature endotherm was most pronounced for flexible film, consistent with its highest AG content, which introduces hydrophilic polysaccharides that retain greater amounts of bound moisture (Zhang et al., 2024). On the other hand, rigid film demonstrated the weakest moisture-loss response, reflecting its higher CA content and more compact microstructure along with fewer water-binding sites.

A broad endothermic transition was observed in the range of 130 $^{\circ}\text{C}$ –170 $^{\circ}\text{C}$. This feature cannot be attributed solely to the glass transition (T_g), as it is accompanied by mass loss. Instead, this region likely represents overlapping thermal events, including volatilization of bound moisture and low-molecular-weight components (such as EO fractions), along with polymer relaxation phenomena (Liu et al., 2024). The depth and position of this transition varied distinctly among the film samples. Flexible film revealed the deepest and lowest-temperature transition at 148 $^{\circ}\text{C}$, indicating the greatest reduction in

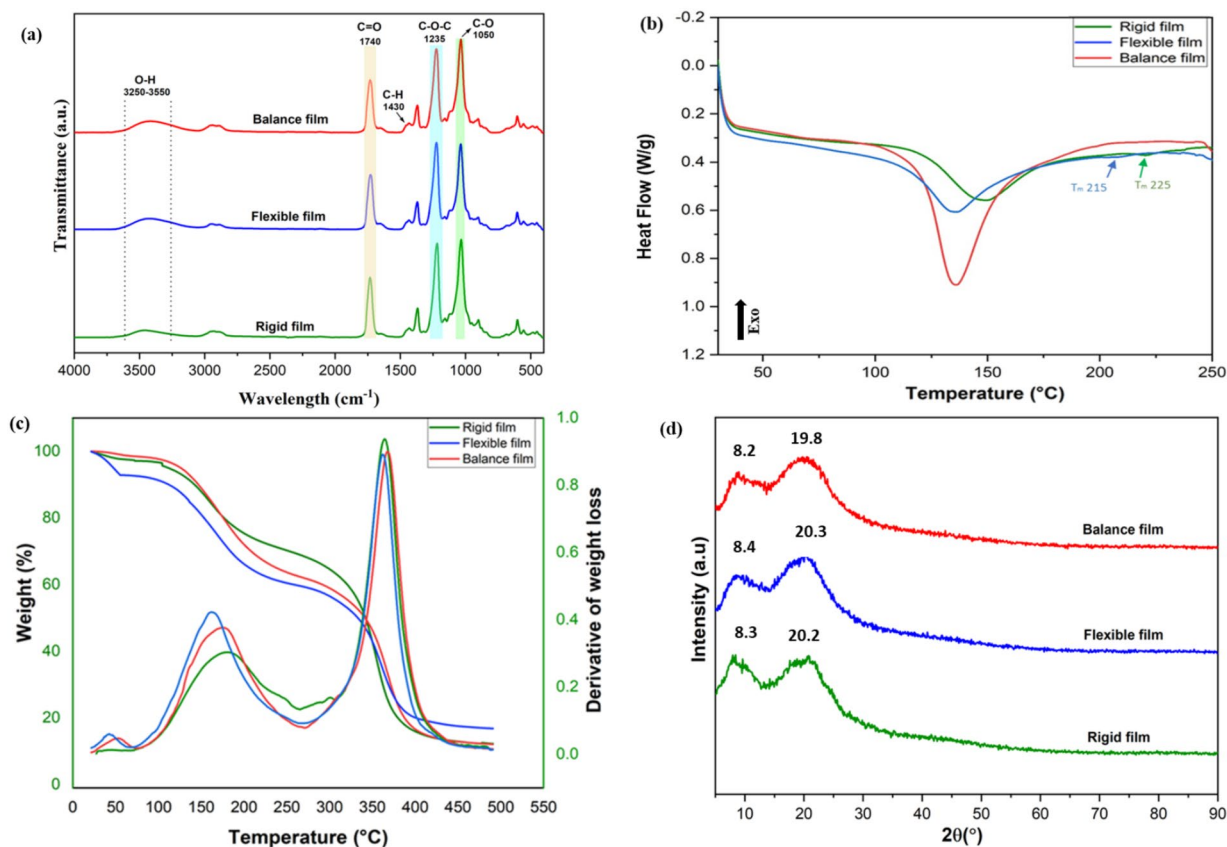


FIGURE 2

(a) FTIR spectra illustrating the functional groups of CA-AG-EO films; (b) DSC thermograms showing thermal transitions of CA-AG-EO films; (c) TGA curves of the CA-AG-EO films; and (d) XRD patterns illustrating the structural order of CA-AG-EO films.

thermal rigidity due to strong plasticization caused by AG gel. The presence of hydrated regions increases chain mobility and lowers the energy required for segmental motion (Saad et al., 2021). Rigid film exhibited the shallowest transition at 170 °C, demonstrating restricted molecular mobility and stronger polymer-polymer interactions within the rigid matrix of the film. However, balanced film showed an intermediate transition at 165 °C.

At higher temperatures, additional endothermic transitions were observed, associated with thermal softening or melting-like relaxation rather than actual crystalline melting, since CA is mainly amorphous or semi-amorphous in nature (Dhongade et al., 2025; Dreux et al., 2019). Balanced film displayed a small and a weak endothermic peak around 210 °C–220 °C, which can be due to the localized chain relaxation or partial disruption of weakly ordered regions within the plasticized matrix of the film. In contrast, rigid film showed a more pronounced high-temperature endothermic response at temperatures around 225 °C, showing enhanced thermal relaxation supported by the high CA concentration (Dhongade et al., 2025). The EO acts as a secondary plasticizer, facilitating molecular rearrangement at elevated temperatures while also maintaining structural coherence. Moreover, flexible film displayed only a subtle high-temperature response, further confirming its greater thermal stability and higher degree of plasticization.

Overall, the DSC results demonstrate that AG is primarily responsible for moisture retention and glass-transition behavior of the films, while compositional differences determine the extent of high-temperature thermal relaxation. Rigid film exhibited a more pronounced high-temperature endothermic response, indicating enhanced thermal relaxation within the CA-rich matrix. Furthermore, the balanced film displayed a small and weak melting-like endothermic peak, indicating localized chain relaxation within the plasticized matrix.

3.1.4 Thermogravimetric analysis

The TGA was carried out to assess the thermal stability and degradation behavior of the selected CA-AG-EO films, and the TGA curves are shown in Figure 2c. The key degradation temperatures, including T10, T20, T50, maximum degradation temperature (Tmax), and the residual mass at high temperature, are summarized in Table 1.

All samples exhibited an initial weight-loss stage in the temperature region (approximately 30 °C–70 °C). This loss is due to the evaporation of physically adsorbed and loosely bound moisture as well as the release of low-molecular-weight volatile species. This is a common phenomenon for electrospun fiber mats due to their large surface area and ability to physically retain a small amount of moisture and volatile additives (Niiniivaara and Cranston, 2020). Among the samples, the flexible film showed had the lowest T10 (105.4 °C), indicating an early onset of mass loss. This could be due to the high AG content and higher hydrophilicity. However, the rigid film (142.4 °C) and the balanced film (145.9 °C) demonstrated higher T10 values, suggesting lower sensitivity to moisture and volatile content. These variations are due to differences in the amount of physically bound moisture and volatiles, rather than film polymer chemistry.

This trend was also observed for the temperatures at which the weight loss reached T20 (148.2, 176.8, and 174.3 °C for the flexible, rigid and balanced film, respectively). In this temperature range, the observed weight loss is due to the continued release or volatilization of physically trapped AG components and EO fractions, rather than the onset of CA decomposition. Accordingly, T10 and T20 values

TABLE 1 Thermogravimetric data of CA-AG-EO films.

Film type	T10 (°C)	T20 (°C)	T50 (°C)	Tmax (°C)	Residue at 500°C (%)
Rigid	142.4	176.8	342	364.4	1.11
Flexible	105.4	148.2	315.2	362.2	0.78
Balanced	145.9	174.3	337.4	368.1	0.84

mainly reflect the thermal evolution of volatile components within the electrospun mat. The earlier onset degradation (lower T10 and T20) observed in the flexible film is due to its higher AG content, which incorporates hydrophilic and thermally less stable components into the matrix. These components enhance moisture retention and include low molecular weight compounds that volatilize at lower temperatures (Zhao et al., 2023). The temperature corresponding to T50 provides a more accurate indicator of the bulk thermal stability of the polymer matrix associated with the main decomposition stage. The highest T50 value (342.0 °C) was observed in the rigid film, followed by the balanced film (337.4 °C), while the lowest value (315.2 °C) was observed in the flexible film. The lower T50 observed in the flexible film indicates a faster progression towards the main decomposition process, which is impacted by the presence of a higher fraction of thermally less stable bioactive components and their influence on the mass-transfer and heat-transfer behavior during heating.

Tmax of films were observed in a narrow range of approximately 362 °C–368 °C for all samples. This degradation corresponds to the primary thermal decomposition of the CA backbone, including chain scission and deacetylation reactions, as commonly reported for CA-based materials (Dreux et al., 2019). Balanced film exhibited the highest Tmax (368.1 °C), followed by rigid film (364.4 °C), while flexible film showed a slightly lower Tmax (362.2 °C). The relatively small differences in Tmax indicate that the incorporation of AG and EO does not significantly alter the fundamental degradation mechanism of CA. At high temperatures, all films exhibited low residual mass, confirming near-complete thermal decomposition of the organic components. Rigid films showed marginally higher residue compared to balanced films and flexible films, which can be attributed to its higher CA content. Overall, the TGA results demonstrate that all CA-AG-EO films follow comparable thermal degradation pathways dominated by the intrinsic thermal behavior of CA. Variations in low-temperature mass loss are primarily associated with differences in moisture and volatile content arising from AG and EO incorporation, while the main degradation temperatures remain largely unaffected.

3.1.5 X-ray diffraction

XRD was performed to evaluate changes in structural order and crystallinity of the selected CA-AG-EO films. The diffraction patterns are presented in Figure 2d. All samples exhibited broad diffraction halos in the range of $2\theta \approx 18$ – 20 , arising from short-range amorphous interchain packing characteristic of organic polymers including CA (Borges et al., 2023; de Freitas et al., 2017). Specifically, the main amorphous halo appeared at approximately 20.2° for rigid film, $\sim 20.3^\circ$ for flexible film, and $\sim 19.8^\circ$ for balanced film, these minor variations in halo position are within the expected range for amorphous electrospun polymeric systems indicating small compositional and processing-related heterogeneities inherent to electrospun fibrous mats. In

addition, all samples exhibited a weak and broad low angle ($2\theta \approx 8-10^\circ$) diffraction feature. This feature has been commonly reported for acetylated cellulose systems and is generally attributed to short-range intermolecular spacing or local ordering effects. The presence of this low-angle feature across all CA-AG-EO formulations indicates the existence of limited short-range order while consistent with the suppression of long-range crystalline organization due to acetyl substitution, which disrupts the native microfibrillar structure of cellulose (Ovando-Roblero et al., 2025).

However, both features were smoother and less defined than those typically reported for neat CA in the literature, suggesting that AG and EO modified the local packing of CA chains. While AG itself exhibits amorphous XRD pattern, it may have contributed to comparative smoothening of the peaks as compared to neat CA in literature (Abu-Thabit et al., 2026; Ekambaram and Dharmalingam, 2020). This is further illustrated by a comparatively smoother halos in flexible film which had comparatively higher AG. Similarly, EO likely contributed to a plasticizing effect, leading to a more disordered amorphous network (Abdalla et al., 2023). Also, the incorporation of AG and EO did not induce new crystalline phases within the CA matrix. The XRD results are consistent with the DSC results, which showed a broad glass transition and the absence of a well-defined melting peak for all CA-AG-EO films. These results indicate that the films lack long-range crystalline order and that their thermal behavior is governed by amorphous chain dynamics rather than crystalline melting.

3.2 Physical and water barrier characterization

3.2.1 Moisture loss, solubility, and swelling index

The moisture loss, solubility, and swelling behavior of the selected CA-AG-EO films are summarized in Table 2, and the observation varied significantly depending on the composition of the film for the moisture loss. The flexible film showed the highest moisture loss, approximately 33%, which is likely due to its high AG content. AG is hydrophilic, rich in polysaccharides with abundant hydroxyl groups, so it tightly binds water molecules that resulted in retained moisture capacity (Zhang et al., 2022). Whereas the rigid film exhibited the loss around 15%, and showed the lowest moisture loss, which is probably due to the higher proportion of CA and denser polymer network with fewer hydrophilic sites for water interaction. The balance film however displayed a moderate moisture loss of roughly 22%, aligning with its balanced formulation. These patterns align with what was observed in the thermal analysis, particularly the early weight loss region below 100 °C in TGA and the low-temperature moisture-loss events in DSC.

A similar concentration driven pattern has been observed for solubility of the film. The flexible film showed the maximum solubility of approximately 34%, which suggests that increased polymer-water

interactions and partial dissolution of the matrix by the strong plasticizing effect of AG. The hydrated regions seem to facilitate water penetration, and act to disrupt the polymer network, which explains the increase in solubility of the film. In contrast, rigid film showed the lowest solubility (about 14%) due to stronger CA-CA intermolecular interactions. Balanced film again demonstrated intermediate solubility (around 23%) because of balance between structural integrity and controlled plasticization. The CA-rich film's lower solubility is also aligned with its higher thermal stability as shown in DSC and delayed degradation as shown in TGA.

The swelling index of the CA-AG-EO films showed negative values for all formulations, ranging from around -7 to -26% , suggesting that rather than the films swelling upon exposure to water, they showed a net mass loss upon immersion in water. This is not a shrinkage effect, but it can be explained by the fact that certain components of the films, which may be bound loosely or are water soluble, leached out during immersion, thus producing a mass loss associated with degradation, rather than true volumetric swelling.

The highest negative swelling index was recorded for the flexible film with the largest mass loss of about -25% , which may be due to the high AG content. AG is a highly hydrophilic polymer with water-soluble polysaccharides, which leach in water. The considerable mass loss may be attributed to the weakening of the polymer matrix due to the high content of AG, which led to the release and diffusion of soluble components. Conversely, rigid film showed lowest mass loss (swelling index below -10%), suggesting better water resistance and superior structural stability in water. This is linked to its higher CA content that provides water resistance, as a result of hydrophobic acetyl groups. Balanced film, with mass loss (-15%), has a moderate effect of AG-induced water sensitivity and EO-induced hydrophobicity, which prevents excessive leaching, indicating a balanced effect of both bioactive compounds. Overall, the swelling index results underline the dual role of AG and EO in determining the stability in water. AG increases water sensitivity and facilitates mass loss (leaching) while EO is a hydrophobic stabilizing agent that reduces moisture penetration and preserving matrix integrity when present at sufficient levels.

3.2.2 Contact angle

Surface hydrophilicity of the CA-AG-EO films was assessed using static water contact angle measurements, and the values are presented in Table 2. All films demonstrate hydrophilic characteristics with the contact angles vary from about 11.60° to 47.59° . The rigid film shows the highest contact angle, close to 47.59° , and hence has the least hydrophilic surface. This observation is consistent with its high CA concentration, as the presence of acetyl groups reduced the surface polarity and limit the diffusion of water from film surface (Boчек and Kalyuzhnaya, 2002). The SEM images, showing a denser and more compact surface morphology, further support this interpretation.

TABLE 2 Functional and physical properties of CA-AG-EO films.

Film type	Moisture loss (%)	Solubility (%)	Swelling index (%)	Contact angle ($^\circ$)	Water permeability ($\times 10^{-10}$), g·m/($\text{m}^2\cdot\text{Pa}\cdot\text{s}$)
Rigid	14.86 ± 1.40^c	14.08 ± 0.12^c	-6.16 ± 2.19^a	47.59 ± 0.51^a	4.07 ± 2.12^b
Flexible	33.26 ± 2.0^a	33.92 ± 2.62^a	-25.61 ± 5.27^c	19.18 ± 0.16^b	4.57 ± 1.20^a
Balanced	21.68 ± 0.80^b	23.10 ± 2.5^b	-15.40 ± 0.85^b	11.60 ± 0.38^c	4.61 ± 4.24^a

Different superscript letters shows significant difference ($p < 0.05$) between different film samples.

On the other hand, the flexible film demonstrated a markedly lower contact angle, dropping to around 19.18° , indicative of high surface hydrophilicity. This behavior is probably linked to its high AG gel content, which promotes the presence of hydroxyl-rich polysaccharides on the film surface thus reducing the contact angle (Zhang et al., 2022). The presence of hydrophilic functional groups enhances water-surface affinity, promoting rapid droplet spreading. This improved wettability is consistent with the elevated moisture loss, solubility, swelling-related mass loss, and water vapor permeability of the film.

In comparison, the balanced film shows the lowest contact angle, around 11.60° , making it the most hydrophilic among all three samples. Despite considering its higher EO content, which might be expected to introduce some hydrophobic character. It suggests that surface behavior is not controlled by EO alone. The combination of moderate AG content and possible surface rearrangement might be exposing more polar groups or even increasing surface roughness, which can enhance water dispersion.

3.2.3 Water vapor permeability

The WVP values and the corresponding values of selected CA-AG-EO films are presented in Table 2. The differences in WVP values among the selected films are evident, reflecting the impact of formulation variables on moisture diffusion properties. The rigid film exhibited the lowest WVP value of about 4.07×10^{-10} g·m/(m²·Pa·s) among the selected films. This observation is consistent with high CA content (5.7%) which in turn reduces the permeability and leads to dense and compact polymer matrix. CA's acetyl groups, which are hydrophobic, reduces the interaction with water, and suppress water vapor diffusion by decreasing the free volume and restricted permeation pathways (Bochek and Kalyuzhnaya, 2002). This behavior agrees with the low moisture loss and swelling in the rigid film. On the other hand, the flexible and balanced films were statistically similar ($p > 0.05$) but higher than rigid film. Both had comparatively lower content of CA content, with the balanced film having the least (4.2%). Despite having 5.1% CA in flexible film, its high AG content, which incorporates hydrophilic polysaccharide and provides more free volume within the matrix, increasing the water vapor mobility (Hadi et al., 2023).

3.3 Color attributes and UV–visible optical properties

CA-AG-EO films have shown different visual appearance depending on the film's composition. These optical properties are advantageous for food packaging applications. Opaque films allow effective protection of light-sensitive products against ultraviolet and visible light, whereas transparent films enhance visual appeal and consumer appeal. Changes in ΔE and lightness (L^*) also affect product appearance and quality.

The lowest L^* value was obtained for the flexible film, indicating a relatively darker appearance (Figure 3a). On the other hand, the balanced film had a higher L^* , suggesting it is whiter than the other films. This can be attributed to its higher EO content, which might induce the formation of EO-rich domains in the CA-AG matrix (How et al., 2024). The rigid film showed an intermediate L^* value, which agrees with its composition and low EO content. The a^* values of all

the films were close to zero, suggesting that the films exhibited no significant red or green tones. Likewise, b^* values were small, suggesting that there was little yellow or blue tint to any of the formulations. These findings suggest that the incorporation of AG and EO had most impact on the opacity and brightness of the films, rather than their color.

The ΔE values of the different films varied considerably (Figure 3b). The flexible film presented the highest ΔE (88.20 ± 0.55), mainly due to its low L^* value, which corresponds to a darker appearance and thus a greater difference from the reference sample. The rigid film had intermediate ΔE values (74.90 ± 0.15) consistent with its intermediate lightness. On the other hand, the balanced film showed the lowest ΔE (56.29 ± 0.30), which consistent with its higher L^* and lighter, whiter appearance. From these findings, it can be concluded that the differences in ΔE between the films are primarily controlled by the L^* (lightness) component and not by variations in a^* or b^* coordinates.

The CA-AG-EO films' optical transparency (600 nm) measurements agreed with the visual inspection and colorimetric results (Figure 3c). The balanced film demonstrated the lowest transparency (0.62 ± 0.044 AU mm⁻¹), which translates to the highest opacity. This translates to a higher intrinsic scattering of light due to the higher EO content. By comparison, the flexible film had the highest transparency (10.55 ± 0 AU mm⁻¹), reflecting its relative lack of opacity and more uniform structure. The rigid film had intermediate transparency (3.92 ± 0.12 AU mm⁻¹), due to its intermediate structure. The higher opacity of the EO-rich films is probably a result of the increased structural heterogeneities and refractive index mismatch between the EO-rich regions and the rest of the polymer matrix, which increases light scattering (Adame et al., 2025).

The UV–visible permeability of CA-AG-EO films is presented in Figure 3d. All films showed good UV absorption in the UV region (300–800 nm), demonstrating good UV blocking. In the visible region (400–800 nm) the spectra were fairly flat, with no significant differences observed among different formulations. The balanced film had the lowest permeability in the visible range, which translates to the highest opacity. The flexible film, on the other hand, had the highest permeability, suggesting the best optical transparency among the samples. The rigid film had an intermediate value. The balance film exhibits low transparency due to the presence of hydrophilic and amorphous polysaccharide regions introduced by AG, which disrupt the uniformity of the polymer matrix and increase light scattering, thereby enhancing opacity (Pattanayak et al., 2025). This enhanced transparency in the flexible film is probably due to the combined plasticizing effect of EO and AG, which enhances the compatibility between phases and decreases the heterogeneity of the matrix, reducing light scattering. This order of transparency agrees with opacity measurements, visual appearance and microstructural analysis.

3.4 Antioxidant and antimicrobial properties

The antioxidant capacity of the CA-AG-EO films was evaluated using ABTS and DPPH radical scavenging assays. All the three film formulations demonstrated antioxidant activity, with distinct differences in performance mainly linked to the concentrations of AG and EO used during their fabrication. These findings confirm the retention of bioactive compounds within the matrix and highlight their functional contribution to radical scavenging. Among all samples, balanced film exhibited the highest antioxidant activity in both ABTS

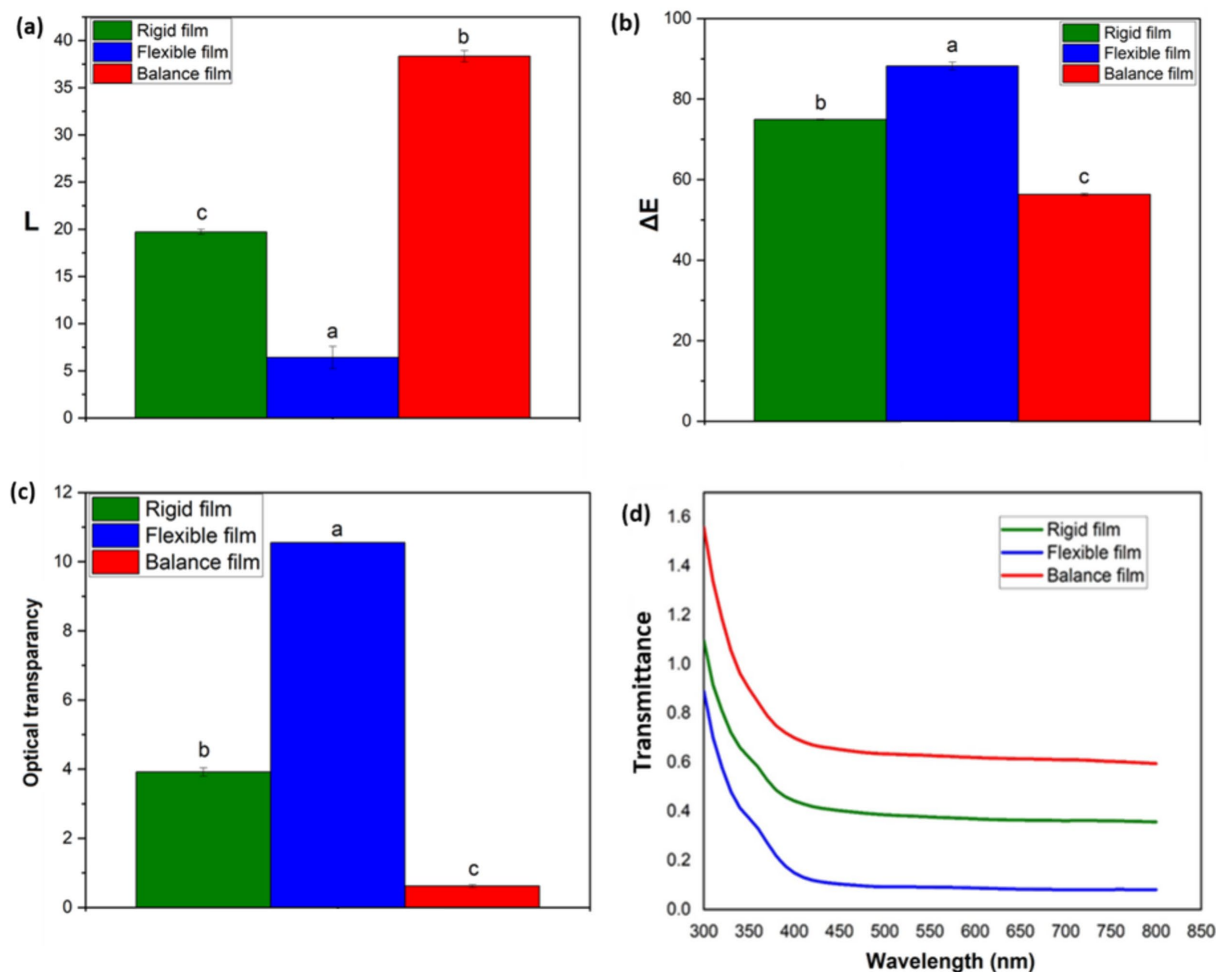


FIGURE 3

Color and optical parameter of CA-AG-EO films: (a) L^* , (b) ΔE , (c) Optical transparency, and (d) UV-visible absorbance.

($318.40 \pm 7.33 \mu\text{g GAE/g film}$) and DPPH ($300.42 \pm 16.47 \mu\text{g GAE/g film}$) assays. The superior performance of this formulation is attributed to the high EO concentration (1.5%). These volatile constituents remained active after electrospinning, confirming the method's effectiveness in preserving sensitive essential oil components.

Flexible film, with a high AG content (25%), showed antioxidant capacity (ABTS: $232.76 \pm 12.25 \mu\text{g GAE/g film}$, DPPH: $143.94 \pm 16.92 \mu\text{g GAE/g film}$). Given that AG is primarily aqueous in nature, it is possible that a portion of its antioxidant constituents may not be fully accessible during extraction, likely due to physical entrapment within the electrospun matrix (Mailley et al., 2021). This may involve partial depletion of water-soluble bioactives and reduced retention within the fiber matrix. The lowest antioxidant activity was observed in rigid film (ABTS: $195.54 \pm 4.83 \mu\text{g GAE/g film}$, DPPH: $103.32 \pm 10.23 \mu\text{g GAE/g film}$), which contained the lowest EO concentration (0.3%). The reduced performance may result from insufficient EO to provide meaningful radical scavenging and limited contribution from AG alone at this level.

The antibacterial activity of CA-AG-EO films was assessed using the agar disk diffusion method against two Gram-positive strains, *Bacillus cereus* and *Staphylococcus aureus*, and two Gram-negative strains, *Salmonella enterica* and *Escherichia coli*. The results revealed clear differences in antimicrobial efficacy based on film composition, particularly the concentration of EO, which is known for its potent

antimicrobial properties (Rabbani et al., 2026). Among all the samples, the balanced film showed the most effective and robust antibacterial activity with clear zones of inhibition against all four bacterial strains. *Escherichia coli* ($10.8 \pm 0.7 \text{ mm}$) had the highest zone followed by *Bacillus cereus* ($9.16 \pm 0.90 \text{ mm}$), *Staphylococcus aureus* ($8.3 \pm 0.7 \text{ mm}$) and *Salmonella enterica* ($7.8 \pm 0.62 \text{ mm}$). This was due to the high concentration of EO incorporated in the film, which offers a rich source of active terpenoids that can damage the bacterial membrane, and thus hinder bacterial growth. Flexible films had selective antibacterial properties. The zones of inhibition were observed for *Bacillus cereus* ($5.73 \pm 0.15 \text{ mm}$), *Salmonella enterica* ($6 \pm 0.1 \text{ mm}$) and *E. coli* ($6.3 \pm 0.2 \text{ mm}$) but not for *S. aureus*. The lack of activity against *S. aureus*, lower zones of inhibition, and lower EO concentration may be explained by the loss of EO during electrospinning, because its volatile components were lost due to rapid solvent evaporation under the influence of a high electric voltage (da Cruz et al., 2024). On the other hand, the rigid film showed no effect against any of the strains. This indicates that a lower concentration of EO and AG cannot induce antibacterial activity under these conditions. Overall, the results show that antibacterial activity is primarily due to the EO concentration. These results are consistent with other studies that showed the high level of antimicrobial activity of EO is due to membrane and metabolic disruption (Daeialiakbar et al., 2025; Istiqomah et al., 2022; Singh et al., 2023).

4 Conclusion

Three CA-AG-EO films, representing rigid, flexible, and balanced mechanical types, exhibited markedly different structure–property–function profiles, primarily determined by their composition. The CA-rich rigid film formed a compact and thermally stable matrix, with the lowest moisture absorption and water vapor permeability; however, the flexibility of this film was the lowest. The AG-rich flexible film exhibited greater chain mobility, higher moisture loss, solubility, and permeability, indicating its more hydrophilic and plasticized character. The EO-enriched balanced film showed intermediate mechanical and barrier properties, and the superior antioxidant and antibacterial properties, indicating effective incorporation and retention of essential oil components. Among the formulations examined, the balanced film offers the most attractive compromise between mechanical integrity, moisture response, and biofunctional performance.

These results show that a feasible approach to control morphology, thermal stability, water sensitivity, optical properties, and bioactivity within a single material platform can be achieved by adjusting the relative amounts of CA, AG, and EO. These findings position CA-AG-EO electrospun films as promising candidates for active, biodegradable food-packaging applications, alternative to petroleum-based packaging. Further validation under real food systems and long-term storage conditions would further elaborate the feasibility.

Data availability statement

The data analyzed in this study is subject to the following licenses/restrictions: the data that support the findings of this study are available from the corresponding author upon reasonable request. Requests to access these datasets should be directed to Akmal Nazir, akmal.nazir@uaeu.ac.ae.

Author contributions

AR: Investigation, Methodology, Visualization, Writing – original draft. ANi: Methodology, Writing – review & editing. MI: Resources,

Writing – review & editing. MJ: Resources, Writing – review & editing. ANa: Conceptualization, Funding acquisition, Methodology, Resources, Supervision, Writing – review & editing.

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Conflict of interest

The author(s) declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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