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Extraction and applications of frankincense oleoresin as functional ingredient in pectin/sodium-alginate composite films for active packaging

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ABSTRACT

The composite films fabricated by incorporating frankincense oleoresin (FOR) into pectin and sodium alginate matrices, were underwent several analysis including thickness, mechanical strength, barrier properties, color attributes, morphological structure, surface roughness, chemical composition, crystalline nature, as well as evaluations of antioxidant and antimicrobial properties. The findings of the study indicated an increase in the thickness of the film samples with the addition of FOR however reduction in both tensile strength (from 9.59 to 2.58 MPa) and elongation at break (from 9.85 % to 3.22 %) was observed. The water vapor permeability of the developed films demonstrated an increase from 0.411 to 0.878 (g*mm)/(m^2*h*kPa). Atomic Force Microscopy (AFM) imaging results indicated a reduction in the surface roughness of films upon the addition of FOR. Additionally, Fourier Transform Infrared Spectroscopy (FTIR) spectra unveiled chemical interactions between the film-forming polymers and FOR. Furthermore, a significant enhancement in the antioxidant activity was observed in films incorporating FOR, compared to the control group. These FOR-loaded film samples demonstrated antimicrobial effects against *Pseudomonas aeruginosa*, while no inhibitory zones were observed against *Staphylococcus aureus* and *Candida albicans*. In summary, the outcomes of this investigation highlight the promising potential of pectin/sodium alginate composite films containing FOR in packaging applications.

1. Introduction

Over the past few decades, the extensive manufacturing of plastic materials has led to a significant environmental challenge known as "white pollution." This issue arises from the widespread use of non-biodegradable plastic packaging, exerting considerable strain on the global environment. Therefore, the increasing demand for sustainable and environmentally friendly packaging materials has intensified recent studies on the development of active films as an alternative to plastics. This biodegradable packaging can reduce or prevent the spoilage of food products and extend their shelf life while maintaining their sensory and safety attributes (Huang et al., 2019; Zhang et al., 2023).

Among various biopolymers, the films prepared from polysaccharides, such as pectin and sodium alginate (SA) have shown ideal film-forming properties. Pectin is a linear homopolymer, consisting of D-galacturonic acid units chemically linked by α-1,4 glycosidic linkages. This water-soluble, biodegradable, and non-toxic substance exists in the intercellular layer of plants and is mostly extracted from apple pomace,

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citrus peel, and beet pulp. SA is the sodium salt of alginic acid, a linear polysaccharide derived from brown algae, belonging to the Phaeophyceae family, which is polymerized by β-D-mannuronic acid (M) and α-L-guluronic acid (G) through α-1,4 glycosidic linkages. The sequential organization of M and G along the chain, as well as the M/G ratio determine the gelling and film-forming properties of alginate (Gohil, 2011). Despite the great advantages of these biopolymers, their poor mechanical and barrier properties limit their further application in the packaging industry (Marangoni Júnior et al., 2022). To overcome these challenges, researchers have explored various strategies to enhance the functionality of single-layer films. Among these approaches, the combination of two or more biopolymers, through blending, cross-linking, or multilayer formation and the incorporation of different active agents has shown great promise.

Active or intelligent food packaging can be obtained by incorporating synthetic and natural antioxidant/antimicrobial agents into edible films or directly coating them on food products. However, the active agents produced from natural sources are relatively safer and easily obtained, which has attracted much more attention (Bierhalz et al., 2012; Desam et al., 2019; El-Mesallamy et al., 2012). Frankincense, also known as Olibanum, is the oleoresin of Boswellia trees, which has been used since ancient times in India, Yemen, Somalia, and southern Oman to treat wounds and skin infections, dementia, and inflammatory diseases (Byler et al., 2018). The biological properties of Frankincense oleoresin (FOR) have been attributed to its essential oils, as well as its non-volatile diterpenoids and triterpenoids. Incorporation of frankincense oleoresin (FOR) extract into biopolymer-based films can enhance mechanical, barrier and active properties. The chemical components in FOR, particularly boswellic acids and terpenoids, are expected to interact with the film matrix, potentially improving film physicochemical properties. Yang, Jeon (Yang et al., 2010) represented the potential antioxidant activity of FOR due to the presence of high amounts of p-menth-2-en-ol (34.5 %) in its essential oil. Hence, the objective of this investigation was to fabricate composite films with antioxidant and antimicrobial properties by incorporating pectin, SA, and FOR. Furthermore, the study included an assessment of the influence of FOR on the physical, structural, mechanical, and thermal properties of the films.

2. Materials and methodology

2.1. Materials

Pectinand sodium alginate (SA)were sourced from Sisco Research Laboratories (SRL) Pvt. Ltd., based in Mumbai, India. Glycerol was obtained from BDH Laboratory Dorset, UK. All additional chemicals utilized in this study were procured from Sigma-Aldrich, located in St. Louis, MO, USA, and met analytical grade standards.

2.2. Extraction of frankincense oleoresin (FOR)

The powder resin (1.0 kg) was extracted with ethanol (3.0 L) at 50 °C with continuous stirring (3 times). At the end of each extraction, the solvent was filtered to get a clear solution. Finally, all three extractions were mixed and distilled out by rotary evaporator under reduced pressure to obtain the thick sticky mass i.e. frankincense oleoresin (400 g).

2.3. Films preparation

In this study, 2 g/100 mL pectin and 1 g/100 mL sodium alginate solutions were individually prepared under constant stirring at ambient temperature for 60 min. Thereafter, 0.5 mL/100 mL glycerol was incorporated as a plasticizer into each film-forming solution, which was then subjected to continuous stirring at the same temperature for an additional 30 min. These solutions were subsequently mixed to fabricate composite films, maintaining constant stirring for 1 h. The resulting homogenized mixture was then divided into beakers, labeled as PESA, PSF1, PSF2, and PSF3, respectively. PESA was taken as the control composite film and the PSF1, PSF2, and PSF3 samples were introduced with varying concentrations of frankincense oleoresin as presented in Table 1. FOR was first dissolved in a small volume of ethanol, then emulsified into the film-forming solution under magnetic stirring for 30 min. Detailed compositional data of these formulations are presented in Table 1. For the preparation of each film sample, a 20 ml film-forming solution of the respective beakers was carefully taken and casted into sterile polymeric Petri dishes. Films were dried at 40 °C for 48 h in a dust-free chamber to avoid high-temperature degradation of thermolabile compounds in FOR. Following complete drying, the solidified film samples were gently detached from the Petri dishes and prepared for further analytical assessments.

2.4. Transparency and color properties

Transparency of the biopolymer-derived films is crucial for various purposes. It enables consumers to visually evaluate the product, and the transparency of the contents aids in highlighting the quality, color, and overall attractiveness of the item. Transparency analysis of the film samples (PESA-PESA3) was conducted by using a V-10 plus visible spectrophotometer (ONDA, Padova, Italy) at a wavelength of 550 nm, as per the procedure described by Zhao, Wang (Zhao et al., 2022).

The color properties of edible films (i.e., lightness (L*), redness (a*) and yellowness (b*)) were also determined using Konica Minolta CR-5, Konica Minolta Sensing Americas (Tokyo, Japan). The overall color difference (ΔE) was also obtained following the procedure outlined by Bhatia et al. (Bhatia et al., 2024).

2.5. Morphological properties

The morphological characteristics of the composite films were examined using a scanning electron microscope (SEM, JSM-6510 LA, Jeol, Japan) operating at an accelerating voltage of 20 kV. For the sample preparation, the procedure outlined by Bhatia et al. (Bhatia et al., 2024) was followed.

2.6. Atomic force microscopy

The topographic assessment of the films was carried out using atomic force microscopy (AFM) technology (hpAFM, Nano-Magnetics Instruments). Surface images of the films were captured through tapping mode scanning at room temperature. An ACLA cantilever with a spring constant ranging from 36 to 90 N/m was placed above the sample, and the scan dimensions were set at 10 μm * 10 μm, with the height of the surface relief recorded at a resolution of 256 pixels * 256 pixels. Three scans were conducted for each sample at random locations on the thin film surface. The average roughness (Ra) and Root Mean Square roughness (Rq) values of the films were calculated based on the mean data plane.

2.7. Thickness measurement

Analyzing biopolymer film thickness is crucial for quality control,

Table 1
The compositional ratios of the components used for the film-forming solutions.

Film samples	Pectin (g/100 mL)	Sodium Alginate (g/100 mL)	Glycerol (mL/100 mL)	Frankincense oleoresin (g/100 mL)
PESA	2	1	0.5	Blank
PSF1	2	1	0.5	0.5
PSF2	2	1	0.5	1
PSF3	2	1	0.5	1.5

ensuring uniformity and meeting standards. It influences mechanical, barrier, and optical properties, impacting performance in packaging and biomedical applications. A micrometer Yu-Su 150, (Yu-Su Tools) was used to evaluate the thickness of films following the methodology described by Bhatia et al. (Bhatia et al., 2024).

2.8. Mechanical properties

The analysis of the mechanical properties, such as tensile strength (TS) and elongation at break (EAB) of biopolymer-based films is crucial for assessing their structural integrity and performance. These properties determine the ability of the films to withstand stress and deformation, impacting their suitability for various applications such as packaging. The film samples (PESA-PESA3) underwent an analysis of their mechanical properties utilizing a texture analyzer (TA. XT plus, Stable Micro Systems, Godalming, England) following the ASTM D882 standard procedure (American Society for Testing and Materials. ASTM., 2010).

2.9. Water vapor permeability (WVP) analysis of the films

Water Vapor Permeability (WVP) evaluation of the biopolymer films is vital for effective packaging, ensuring optimal moisture resistance and contributing to sustainability by promoting eco-friendly alternatives in the industry. The WVP of the film samples (PESA-PESA3) was evaluated using the gravimetric method, as described by (Bhatia et al., 2023).

2.10. FT-IR spectroscopy analysis

The FT-IR spectra were acquired using a spectrometer (InfraRed Bruker Tensor 37, Ettlingen, Germany), as per the procedure described by Bhatia et al. (Bhatia et al., 2024).

2.11. X-ray diffraction (XRD) analysis

XRD analysis was conducted using a Bruker D8 Discover apparatus operating at 40 kV following the procedure described in our previous study (Abbas Shah et al., 2024). Scanning of the samples occurred within the 2θ angle spectrum spanning 5–50°, with a scan speed set at 0.500 s/point.

2.12. Antioxidant and antimicrobial analysis

Measuring the antioxidant potential of films is important for preserving product quality and preventing oxidative stress. Antioxidants in the films protect against deterioration, such as color changes by neutralizing free radicals. This ensures the effectiveness of edible films in extending the shelf life of packaged products. The assessment of the antioxidant capacity of films involved the use of ABTS cation and DPPH radical scavenging assay, following the methodologies outlined by (Re et al., 1999), as well as (Brand-Williams et al., 1995). In the DPPH assay, 50 mg of film was mixed with 1.95 mL of ethanolic DPPH solution (adjusted to an absorbance of 0.7 ± 0.2 at 517 nm), vortexed for 30 s, and incubated in the dark for 30 min. For the ABTS assay, 15 mg of each film sample was vortexed with 1.9 mL of a 7 mmol/L ABTS radical solution (prepared with 2.45 mM potassium persulfate), and the absorbance at 734 nm was recorded after 6 min. The results obtained for the antioxidant activity were presented as the percentage of DPPH radical and ABTS cationic free radical inhibition with the average of three replicates.

The assessment of film sample antimicrobial efficacy utilized the agar diffusion method, following the procedures outlined by Seol et al. (Seol et al., 2009) and Matuschek et al. (Matuschek et al., 2014). Film samples were examined for antimicrobial activity against Gram-positive bacteria, specifically *Staphylococcus aureus* (ATCC 25923), Gram-negative bacteria *Pseudomonas aeruginosa* (ATCC 27853), and *Candida albicans* (ATCC 10231), yeast species. Film discs (6 mm) were placed on inoculated Mueller-Hinton agar plates and incubated at 37 °C for 24 h. The diameter of inhibition zones was measured, and results were reported as mean values from three replicates.

2.13. Statistical analysis

To conduct the statistical evaluation, we utilized mean and standard error values derived from three distinct observations conducted independently. An analysis of variance (ANOVA) was performed, and the significance ($p < 0.05$) among the means was determined employing Duncan's multiple range test through SPSS software (Version 26, IBM Company, Chicago, IL, USA).

3. Results and discussion

3.1. Optical and microstructure analysis

In the current study, films were developed using pectin and sodium alginate as base materials, with varying additions of FOR (Fig. 1). Initial observations focused on the visual characteristics of these films revealed that PESA (control) exhibited a rough and even surface. However, the incorporation of FOR significantly more than 1g/100 mL led to a reduction in surface roughness and films showed uniform structure depicted in Fig. 1. Previous research similarly demonstrated that increased concentrations of a plant extract can cause structural irregularities in the resulting films (Lim et al., 2021).

The morphological properties of the developed films, encompassing both their structural and cross-sectional characteristics, are shown in Fig. 1. The control film sample exhibited a homogenous surface with the presence of some particles. The integration of the FOR significantly altered the structure of the films and the films exhibited grainy structure. Such structural change can be primarily attributed to the limited solubility of the extract within the film-forming solution. These structural irregularities can be observed when the extract concentration reaches a certain elevated level, thereby impacting the uniformity and overall structural integrity of the film (Kola, 2020). However, when the concentration of the FOR increased from 1 g/100 mL, the films exhibited uniform and even structure. Various research has also shown a similar impact of incorporating plant extract on the structural properties of the resultant films (Nguyen et al., 2020; Piñeros-Hernandez et al., 2017).

The opacity and color of films developed with FOR were evaluated and results are shown in Table 2. PSF3 film sample loaded with a maximum concentration (1.5 g/100 mL) of FOR showed the highest opacity (5.72 ± 0.18), followed by PSF2, PSF1, and the control sample. The incorporation of FOR significantly increased ($p < 0.05$) the opacity of the films in a concentration-dependent manner. This reduced transparency is likely due to the phenolic compounds in FOR, which block UV and visible light in the pectin and sodium alginate-based films. Similar trends were observed in previous studies, where films containing extracts had lower transparency compared to control samples (Kaewprachu et al., 2017; Yuan et al., 2020). The color parameters of the developed films were also affected by the addition of FOR. While the L and a* values showed minimal variations, there was a notable increase in the b* value, rising from 1.18 ± 0.27 to 2.13 ± 0.18 upon adding FOR. This increase in the b* value indicates a slight increase in the yellowness of the films. The overall color difference in the films ranged from 1.53 ± 0.19 in the control and 2.14 ± 0.26 in the PSF3 containing the highest concentrations of the FOR. The overall color difference observed in the films was between 1.53 ± 0.19 in the control group and 2.14 ± 0.26 in the PSF3 group, which contained the highest amount of FOR. Research has indicated that plant extracts, rich in polyphenols, significantly interact with biopolymers, resulting in notable alterations in color properties (Mir et al., 2018). Previous studies have also shown similar results in which the addition of plant extracts altered the color attributes of the resultant films (Mir et al., 2018; Yuan et al., 2015;

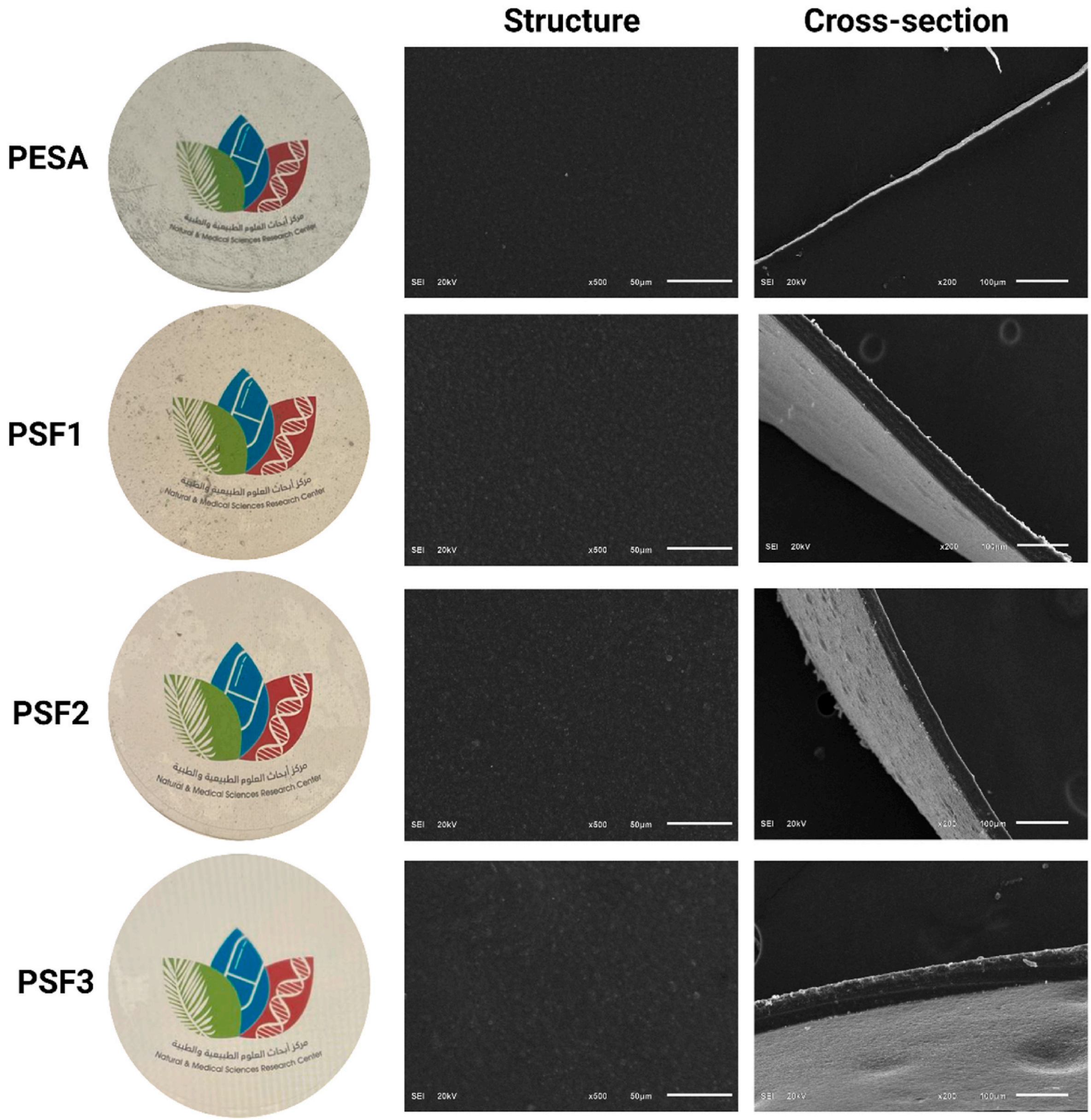


Fig. 1. Visual and SEM analysis of the fabricated films incorporated with FOR. PESA: Pure sodium alginate film; PSF1, PSF2, and PSF3: PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)

Table 2
Opacity and color parameters of the fabricated films.

Sample Codes	Opacity	L	a*	b*	ΔE
PESA	1.81 ± 0.09^{a}	96.96 ± 0.07^{a}	0.04 ± 0.01^{a}	1.18 ± 0.27^{a}	1.53 ± 0.19^{a}
PSF1	2.53 ± 0.20^{b}	97.10 ± 0.05^{a}	0.06 ± 0.02^{a}	0.99 ± 0.13^{a}	1.50 ± 0.09^{a}
PSF2	4.33 ± 0.22^{c}	96.28 ± 0.85^{a}	0.16 ± 0.02^{b}	2.94 ± 0.52^{c}	2.97 ± 0.32^{b}
PSF3	5.72 ± 0.18^{d}	96.54 ± 0.41^{a}	0.17 ± 0.03^{b}	2.13 ± 0.18^{b}	2.14 ± 0.26^{b}

Values are expressed as mean ± SD (n = 3). Values marked with distinct letters (a, b, c, and d) within a column indicate significant differences *($p < 0.05$)*. L: lightness, a*: green-red color, b*: blue-yellow color, ΔE*: overall color variation. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively).

Zhang et al., 2020).

3.2. Atomic force microscopy (AFM)

Atomic Force Microscopy (AFM) images reveal the morphological structure (a qualitative attribute) and surface roughness (a quantitative attribute) of biocomposite films (Hosseini et al., 2015). Fig. 2 displays the surface morphologies and corresponding roughness measurements (Ra and Rq) of the films. The FOR-free PESA film shows a comparably rough surface, with Ra and Rq values of 93.05 nm and 116.4 nm,

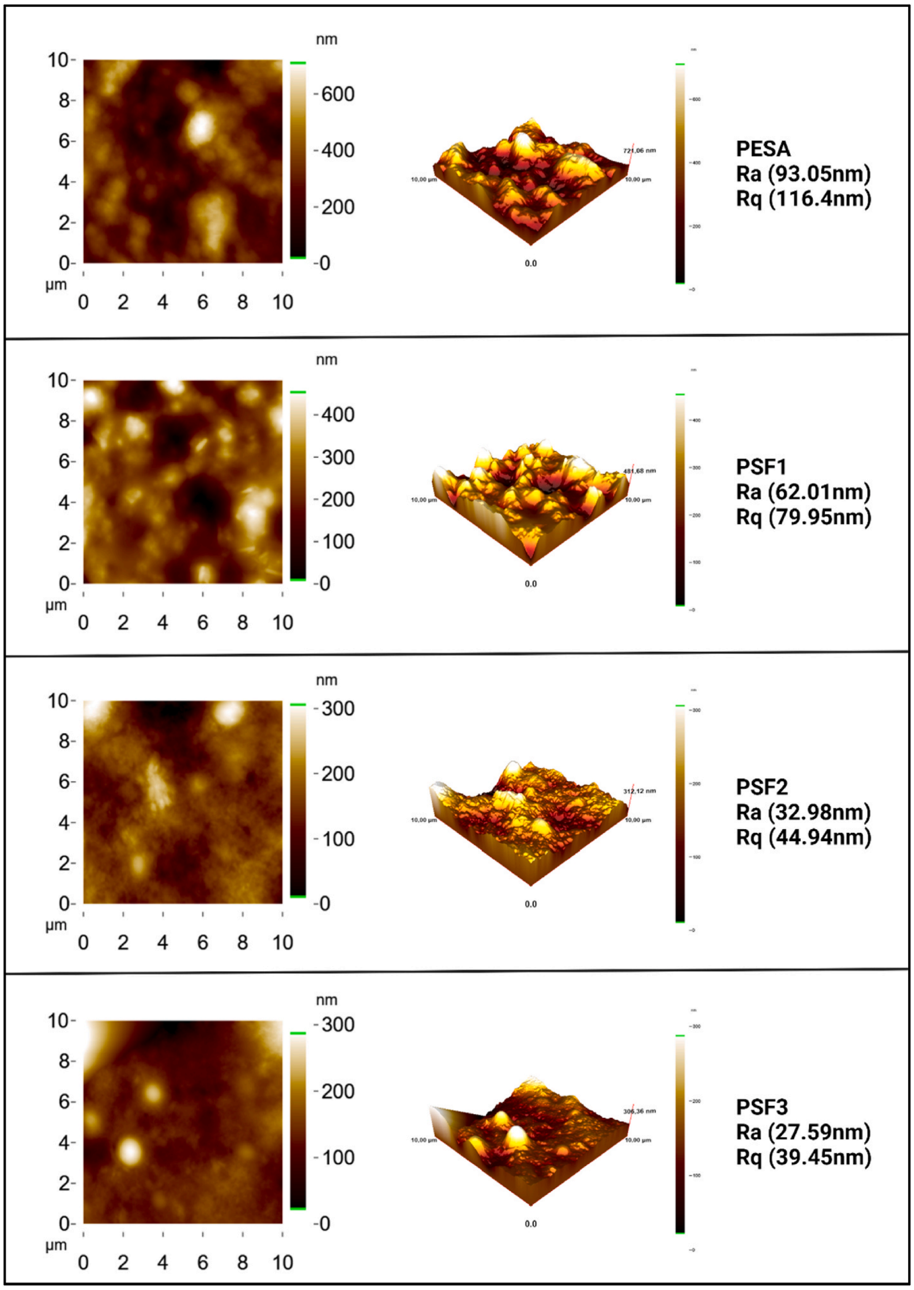


Fig. 2. Atomic Force Microscopy (AFM) images of the film samples. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)

respectively (Fig. 4). Adding FOR significantly reduces film roughness, evident from lower Ra and Rq values. For instance, PSF1 has Ra and Rq values of 62.01 nm and 79.95 nm, respectively; PSF2 shows 32.98 nm and 44.94 nm; and PSF3 has 27.59 nm and 39.45 nm. These findings align with Atarés et al. (Atarés et al., 2010), who noted that soy protein isolate-based films with cinnamon essential oil became smoother due to the liquid oils expanding and filling surface irregularities after drying. Escamilla-García (Escamilla-García et al., 2017) observed similar effects with the addition of various essential oils (anise, orange, cinnamon) to chitosan–zein edible films. An initial decrease in roughness was also noted in studies using *Origanum vulgare* L. essential oil-based composite edible films. However, an increase in roughness was observed with higher oil concentrations, possibly due to lipid aggregation and creaming during drying, leading to surface irregularities (Ghasemlou et al., 2013). The difference in roughness between the control and FOR-loaded films aligns with the SEM microstructural observations.

3.3. Thickness, mechanical and barrier properties of the films

The fabricated films loaded with FOR were analyzed for thickness measurements and the obtained results are presented in Table 3. The thickness of the films increased significantly ($p < 0.05$) with the addition of FOR. The increase in film thickness can be attributed to a higher concentration of solid content incorporated into the film matrix, and to the disruption of its structural arrangement caused by the introduction of the extract (Yuan et al., 2020). The results regarding the TS and EAB for the fabricated films are presented in Table 3. There was a notable decrease in both TS and EAB values with the introduction of the FOR into the films. Specifically, the TS exhibited a significant reduction ($p < 0.05$) from 9.59 ± 0.64 MPa in the control sample (PESA) to 2.58 ± 0.55 MPa in the PSF3 sample, which had the highest FOR concentration. Similarly, the EAB of the PSF3 sample also showed a significant decline ($p < 0.05$) to 3.22 ± 0.30 %, in comparison to the control sample's EAB

Table 3
Thickness, mechanical and barrier properties mean values of film samples. The ± sign mean standard deviations

Film samples	Thickness (mm)	TS (MPa)	EAB (%)	WVP ((g*mm)/(m2*h*kPa))
PESA	0.054 ± 0.005[a]	9.59 ± 0.64[a]	9.85 ± 0.59[a]	0.411 ± 0.012[a]
PSF1	0.070 ± 0.010[b]	5.55 ± 0.44[b]	8.02 ± 0.31[b]	0.489 ± 0.018[b]
PSF2	0.094 ± 0.005[c]	4.45 ± 0.29[c]	3.34 ± 0.07[c]	0.518 ± 0.024[b]
PSF3	0.100 ± 0.010[c]	2.58 ± 0.55[d]	3.22 ± 0.30[c]	0.878 ± 0.030[c]

Values are expressed as mean ± SD (n = 3). Values marked with distinct letters (a, b, c, and d) within a column indicate significant differences *($p < 0.05$)*. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively).

of 9.85 ± 0.59 %. These results suggest that the incorporation of the FOR into the biopolymer film induced structural modifications within the pectin and sodium alginate composite, likely due to the interactions between the biopolymers and FOR molecules (Ju et al., 2020a). Previous studies have also reported similar results in which both TS and EAB of the films decreased with the incorporation of the plant extract (Ju et al., 2020a; Kalkan et al., 2020; Zhang et al., 2020). To improve the mechanical properties, potential strategies such as incorporating cross-linking agents could be employed to enhance the intermolecular bonding within the film matrix.

The results regarding the WVP of the tested films are presented in Table 3. Film samples demonstrated a WVP range of 0.411 ± 0.012 to 0.878 ± 0.030 (g*mm)/(m2*h*kPa). It was observed that the films integrated with FOR exhibited an increase in water permeability. This elevation in WVP may be attributed to the incorporation of FOR, which likely induced heterogeneity and increased porosity within the film matrix (Ju et al., 2020b). Previous research has similarly indicated that incorporating plant extracts into biopolymer-based films impacts their water permeability (Ju et al., 2020b; Nemazifard et al., 2017; Zhang et al., 2020). To reduce water vapor permeability, incorporating hydrophobic components such as natural waxes or essential oils may improve moisture barrier properties. Another approach could involve the development of multilayer films or the use of blended polymers with lower water affinity to hinder water vapor diffusion.

3.4. Fourier Transform Infrared Spectroscopy (FTIR)

The infrared absorption spectrum displayed in Fig. 3 includes the

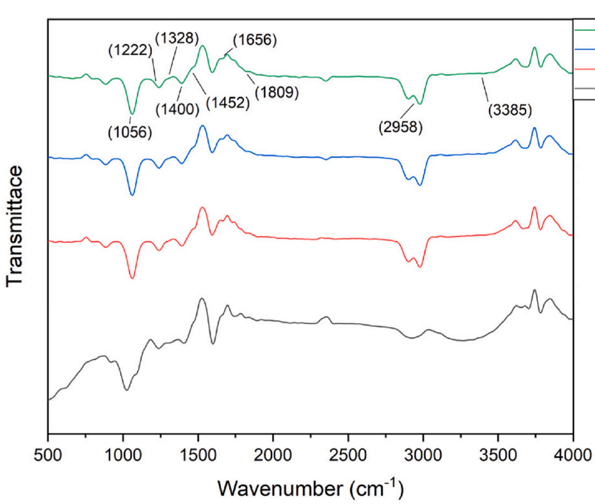


Fig. 3. FTIR overlay of PESA, PSF1, PSF2 and PSF3 films. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)

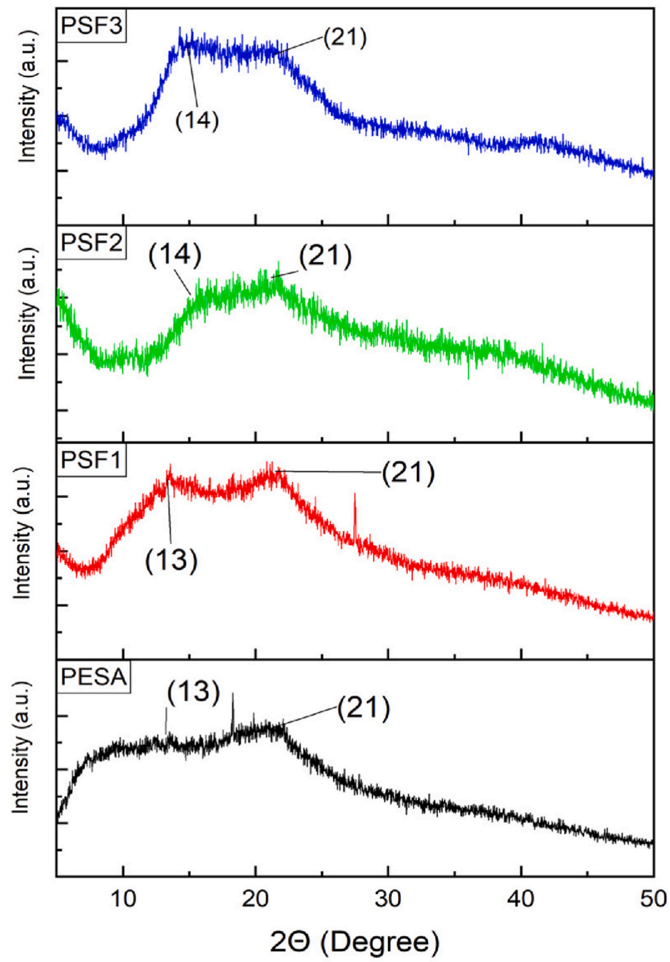


Fig. 4. XRD diffractogram of PESA, PSF1, PSF2 and PSF3 films. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)

distinct peaks of PESA along with PSF1, PSF2, and PSF3. Pectin and sodium alginate composite exhibited specific absorption peaks at 1400 cm^{-1}, which is typical for the symmetrical stretching of the -COO group. The peak at 1056 cm^{-1} corresponds to the stretching of the O-H group of the galacturonic sugar units (Luna-Vital et al., 2018). Additionally, a pronounced and wide peak at approximately 3385 cm^{-1} denotes the collective -OH interactions between molecules (Moni et al., 2021). This broadening is attributed to the hydrogen bonding among the hydroxyl groups (Marangoni Júnior et al., 2022; Snyder et al., 2014).

The FTIR spectral examination of the FOR from *B. sacra* revealed characteristic absorption bands, with the O-H stretching vibration in the range of 3300 cm^{-1}, the aliphatic C-H stretching vibration at 2958 cm^{-1}, the C=O stretching vibration at 1809 cm^{-1}, the C=C stretching vibration at 1656 cm^{-1}, and the C-C stretching vibrations at 1452 cm^{-1} and 1328 cm^{-1} respectively. A study carried out in 2017 by Al-Riyami et al. (Al-Riyami et al., 2017) revealed similar results where the chemical composition of frankincense essential oil was studied.

3.5. X-ray diffraction analysis (XRD)

The X-ray diffraction technique is utilized to analyze the structural composition of polymeric edible films by assessing their crystallinity levels, as well as the dimensions and configuration of the polymer's crystalline regions (Murthy et al., 1990; Siriprom et al., 2014). Fig. 4 presents the XRD diffractogram of the PESA, PSF1, PSF2, and PSF3 edible films. Incorporating FOR extract into the samples resulted in a higher % crystallinity, as evident from the peak intensity. In the present study, the PESA films exhibited two peaks at 13° and 21° of 2θ characteristic of pectin and sodium alginate composite (Fan et al., 2021). The incorporation of extract shifted the peaks slightly to a higher angle of 2θ i.e., 14° was observed in PSF2 and PSF3. The addition of the extract increased the peak intensity and % crystallinity of the films; the control PESA film displayed a % crystallinity of 18.3 %, while a sharp increase in % crystallinity was detected in the film with the highest concentration of FOR (25.6 %). These results suggest that the addition of FOR might be responsible for the increase in the crystalline nature of the composite. The literature revealed that some segments in resins are crystalline which can be the probable reason for the increase in % crystallinity of the PSF film samples (Gashe et al., 2022; Parimal et al., 2011). Additionally, the incorporation of other additives such as essential oils has been reported to increase the crystalline nature of the materials. Hosseini et al. reported that the *Origanum vulgare* L. essential oil (OEO) addition increased the crystallinity of the films (Hosseini et al., 2016).

3.6. Antioxidant and antimicrobial activities of the films

The antioxidative capacity of the film samples was assessed via two distinct methods, namely, the DPPH radical and ABTS cation radical scavenging assays. The corresponding results are depicted in Fig. 5. The control film sample, PESA, exhibited an inhibitory effect of 25.51 % and 18.89 % in the DPPH radical and ABTS cation radical scavenging assays, respectively. Incorporation of FOR into the film samples markedly enhanced the inhibition percentages, elevating them from 25.51 % to 59.89 % in the DPPH assay and from 18.89 % to 40.17 % in the ABTS assay. This notable increase in antioxidative activity upon the integration of FOR can be ascribed to the rich terpene composition of the *Boswellia sacra* extract (Al-Harrasi et al., 2013) (see Fig. 6).

For the antimicrobial analysis, when FOR-loaded films were tested against *Staphylococcus aureus* and *Candida albicans,* no zones of inhibition were observed. The films showed antimicrobial activity against *P. aeruginosa* with the zone of inhibitions as 12.51, 13.43 and 13.93 for PSF1, PSF2 and PSF3 film samples respectively. The antimicrobial efficacy of the film samples was evaluated against three different microorganisms: *P. aeruginosa*, *S. aureus*, and *C. albicans*. In the case of *S. aureus* and *C. albicans*, the films infused with FOR did not demonstrate any zones of inhibition, indicating a lack of antimicrobial activity against these two microorganisms. However, when tested against *P. aeruginosa*, the films exhibited noticeable antimicrobial properties. The zones of inhibition recorded were 12.51 mm, 13.43 mm, and 13.93 mm for the PSF1, PSF2, and PSF3 film samples, respectively. These results indicate that while the FOR-loaded films are effective in inhibiting the growth of *P. aeruginosa*, they do not exhibit similar antimicrobial activity against *S. aureus* and *C. albicans*.

Boswellia sacra, often known as frankincense, has been used for many years as a natural treatment for inflammation and infections caused by microbes in various health conditions (Suther et al., 2022). Its wide-ranging uses are due to its primary active compound, boswellic acids (BAs), which are a type of pentacyclic triterpenic acids (Roy et al., 2016; Siddiqui, 2011). The most effective and widely researched among these BAs is 3-O-acetyl-11-keto-β-boswellic acid (AKBA). Various studies have shown the antimicrobial properties of the frankincense and its extracts. In a previous study, alginate film loaded with frankincense oil exhibited antimicrobial activity against different microorganisms (Saied et al., 2020).

4. Conclusion

In conclusion, the comprehensive characterization of composite films incorporating frankincense oleoresin (FOR) into pectin and sodium alginate matrices has provided valuable insights into their properties and potential applications. The selective antimicrobial activity observed in the films highlights the need for further optimization or combination with other bioactive agents to broaden their effectiveness against a wider range of microorganisms. Future studies can focus on optimizing formulations and processing methods to improve the performance and application of FOR-infused composite films in food packaging. Additionally, testing under real food packaging conditions would provide deeper insights into their applicability for diverse food products, aiding in the scale-up of their industrial use.

CRediT authorship contribution statement

Yasir Abbas Shah: Writing – review & editing, Writing – original draft, Software, Methodology, Conceptualization. **Saurabh Bhatia:** Writing – original draft, Supervision, Conceptualization. **Ahmed Al-Harrasi:** Supervision, Project administration. **Mohammad Tarahi:** Writing – review & editing, Writing – original draft, Data curation. **Muhammad Jawad:** Writing – original draft, Software, Methodology. **Tanveer Alam:** Writing – original draft, Methodology, Formal analysis. **Sevgin Dıblan:** Writing – review & editing, Methodology. **Esra Koca:** Software, Methodology, Data curation. **Levent Yurdaer Aydemir:** Writing – review & editing, Methodology, Formal analysis, Data curation. **Dinu Thomas Thekkuden:** Writing – review & editing, Formal

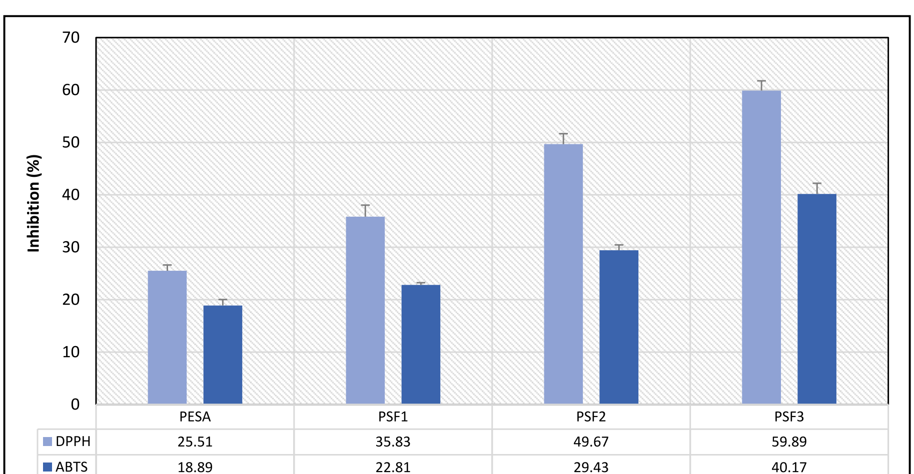
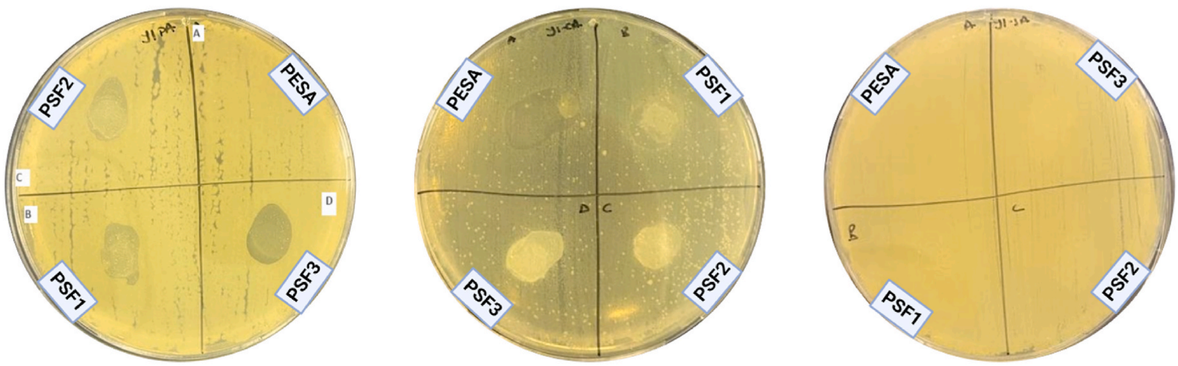


Fig. 5. Antioxidant activity of PESA, PSF1, PSF2 and PSF3 films. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)



Pseudomonas aeruginosa *Candida albicans* *Staphylococcus aureus*

Fig. 6. The results of assessing antimicrobial activity of the films against various microorganisms. PESA: Pure pectin/sodium alginate composite film; PSF1, PSF2, and PSF3: Composite films incorporated with increasing concentrations of frankincense oleoresin (low, medium, and high, respectively)

analysis, Data curation. **Faisal Imam:** Writing – review & editing, Resources, Formal analysis. **Naif Al-Harbi:** Writing – review & editing, Validation, Resources, Formal analysis.

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Declaration of competing interest

Please find enclosed herewith the manuscript entitled "**Extraction and Applications of Frankincense Oleoresin as Functional Ingredient in Pectin/Sodium-Alginate Composite Films for Active Packaging**" for publication in your esteemed journal. There is no conflict of interest among any of the authors whatsoever and all the authors have actively participated in the preparation of manuscript.

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Data availability

Data will be made available on request.

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