Chapter 7
To study the application of the developed films for packaging of food materials

7.1. Introduction

Post-harvest losses of fruits and vegetables remain a critical global challenge, with recent studies indicating that 30-40% of total yield is wasted annually (Shivangi et al., 2021), underscoring the urgent need for improved storage and preservation technologies to enhance food security. This year, the International Potato Day, celebrated annually on May 30th, embraced the theme "Harvesting diversity, feeding hope." In line with this vision, adopting sustainable and active food packaging solutions has become essential to reducing waste and minimizing environmental impacts. The emphasis should be on utilizing eco-friendly materials and environmentally conscious techniques in packaging. Incorporating active ingredients into packaging materials presents a novel strategy for ensuring the consistent release of active agents throughout the entire food supply chain (Dong et al., 2023). Also, our previous chapter has demonstrated the biodegradability of starch/casein and starch and banana-fiber composites incorporating 5% banana fiber (Dutta and Sit, 2024). Biopolymers derived from starch and casein are well-known for their ability to inhibit oxygen through hydrogen-bonded structures effectively and also have displayed a promising ability to incorporate active ingredients, with the added benefit of their affordable cost and impressive processing capabilities and mechanical properties (Dash et al., 2024). However, their hydrophilic properties pose a challenge to their utility as water vapor barriers in food packaging applications. Lipid-based products exhibit excellent resistance to moisture due to their innate hydrophobic properties (Zubair et al., 2021). Also, our previous reports on ultrasound-treated casein films have shown higher emulsion activity (%) and emulsion stability (%) (Dutta and Sit, 2022). The incorporation of active compounds into water-soluble coating solutions enhances their typically poor adhesive properties of fruit and vegetable surfaces that are hydrophobic (Dash et al., 2024). This enhancement is due to the presence of phenolic components within the active compounds, which possess superior adhesive characteristics. However, these films often display increased brittleness and significant thickness, which can result in an aesthetically unpleasing appearance (Shah et al., 2024). However, it might be necessary to further enhance the composite edible films made from plant-based protein, carbohydrate, and lipid sources to improve their tensile strength, barrier qualities, and water resistance. Processes to enhance these properties, like chemical alterations or incorporating nano-reinforcements, need to be done for product preservation (Rawat and Saini, 2024). Protein-carbohydrate-based composite edible film combinations with added lipids are proposed to enhance overall film attributes (Bizymis et al., 2022). The development of enhanced composite films has emerged as a critical focus in addressing the inherent limitations of single-component materials, particularly in improving water barrier properties. While the addition of fillers to polymer matrices can adversely enhance the physical attributes of films, the effectiveness of such improvements is fundamentally dependent on the compatibility and miscibility between the polymer and filler materials (Ghosh et al., 2024).

Recent advances in bio-based packaging substances have demonstrated promising potential in extending food shelf life thereby lessening the impact on the environment. Particularly, the integration of natural antimicrobial compounds and agricultural by-products into biodegradable films has emerged as a sustainable strategy for food preservation (Sukyai et al., 2018). While extensive research exists on bio-based films, the development of active films derived from casein, potato starch, cinnamon, and clove essential oils, reinforced with banana fibers for grape preservation, represents an innovative approach in sustainable food packaging. Through systematic evaluation, this film-forming solution was investigated as a coating material for grapes to assess its preservation efficacy. The significance of this research extends beyond conventional applications, as it addresses both the sustainable use of agricultural by-products and advances the capacity of bio-based films in modern food packaging including preservation systems.

7.2. Materials and methods

7.2.1. Shelf-life study on packaged grapes

For improvement in the shelf-life of packaged items, an active biodegradable film developed (PSC-BPF-CNO) from potato starch (PS)-casein (C) blends, reinforced with banana pseudostem fiber (BPF) and incorporated with 1% cinnamon essential oil (CNO), was evaluated as a protective film for grapes utilizing an altered version of the technique outlined by Shehata et al. (2020). Uniform-sized grapes with flat surfaces were selected and packaged using three distinct materials: the developed active film (PSC-BPF-CNO), fiber-reinforced film (PS-CS-BSF), and commercial packaging material, polypropylene (PP) packaging with the same thickness (0.35 mm) serving as a control. Microbial analysis was conducted using the total plate count method, with results expressed in colony-forming units (CFU/mL). Visual inspection for microbial growth was performed on all packaged samples throughout the storage period. Microbial assessment was carried out at predetermined intervals of 0, 3, 6, 9, and 12 days of storage. The total microbial count plates were incubated at 30℃ for 48 h, while mold and yeast count plates were regulated at 25°C for 5 days. The bacterial load was quantified and expressed as log CFU/g, whereas mold and yeast counts were reported directly as CFU/g.

7.2.2. Study of storage conditions on packaged grapes

7.2.2.1. Total soluble solids (TSS)

Total soluble solids (TSS) measurements in packaged grapes were performed using a digital refractometer at room temperature according to Kumar et al. (2019) protocol. The refractometer was cleaned and calibrated with distilled water before measurements, with results expressed in degrees Brix (°Bx).

7.2.3.2. Titratable acidity (TA) and pH

Titratable acidity (TA) assessment followed a modified version of the Kumar et al. (2019) titration method. The procedure involved homogenizing 10 g of pulp with 90 mL distilled water and adding 3-4 drops of phenolphthalein indicator. This mixture underwent titration against 0.1 M NaOH solution. pH determination followed standard methodology using a digital pH meter, measuring a suspension of 20 g sample in 100 mL purified water, with results reported as average pH values.

7.2.3.2. Physiological loss in weight (PLW)

The amount of weight lost by each packed grape sample, expressed as a percentage, was calculated using a gravimetric method (Basumatary et al., 2022). This was done using equation **7.1**, which incorporates the grapes's initial weight (W_i) and ending weight (W_f). The ending weights were measured after the grapes had been stored for various durations 0, 3, 6, 9, and 12 days.

$$\frac{W_i - W_f}{Wf} \times 100 \qquad \textbf{(7.1)}$$

7.2.3.3. Firmness

Firmness evaluation of packaged grapes during storage utilized a texture analyzer (Stable, Microsystem, UK) following the Basumatary et al. (2022) methodology. The analysis employed a 5 kg load cell with a 2 mm aluminum needle probe operating at 10 mm/min speed, puncturing five equatorial surfaces of fruit samples. Firmness values, reported in Newton (N), represented the mean of three measurements of the force required for fruit surface penetration.

7.2.4. Statistical Analysis

The SPSS statistical software (version 26, SAS Institute Inc., Cary, NC, USA) was used to obtain the experimental data indicated as mean ± standard deviation. One-way (ANOVA) and Duncan's multiple range test (DMRT) with a probability *($p<0.05$)* were taken into consideration for the determination of statistical differences.

7.3. Results and discussions

7.3.1. Application of developed film on packaging of grapes

Fig. 7.1 shows a comparison of microbial growth on three different types of packaging films used for grape preservation over 10 days. The study of microbial growth on Polypropylene (PP) film revealed a progressive increase over time. Observations indicated that by days 8-10, visible colonies had formed on the agar plates, signaling substantial microbial activity. Yellowish spots on the petri plates highlighted significant bacterial proliferation, providing clear evidence of microbial colonization. A similar pattern of microbial growth to PP film has been observed for fiber-reinforced film. The brownish coloration on Day 10 suggests possibly more extensive microbial contamination. In the case of the active film (PSC-BPF-CNO), notably less microbial growth compared to the other two films has been seen. Maintains relatively clear petri plates even up to Day 10 suggesting better antimicrobial properties. The results suggest that the developed active film (PSC-BF-CNO) is most effective at inhibiting microbial growth, likely due to incorporated antimicrobial properties. This would make it potentially better suited for extending the shelf life of grapes compared with standard PP film or fiber-reinforced film (PS-CS-BPF). The increasing colonization seen in the PP and fiber-reinforced films over time is typical of natural microbial spoilage during fruit storage, while the active film (PSC-BPF-CNO) successfully suppresses this microbial growth.

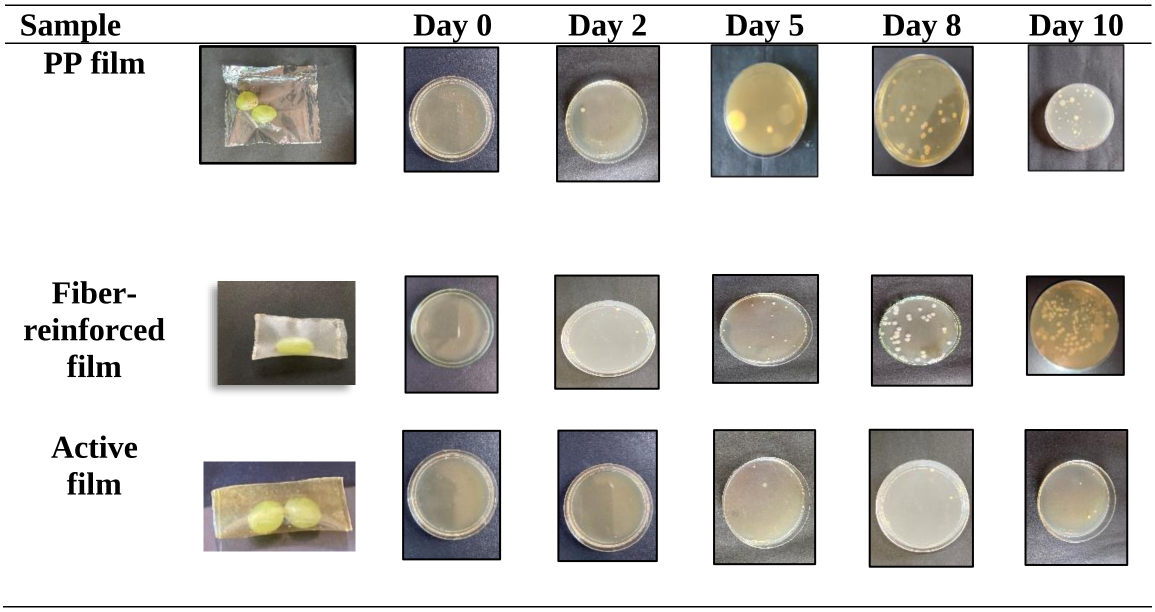

Fig. 7.1. Microbial count after 10 days of storage (5°C) in packaged films

7.3.2. Total soluble solids (TSS)

The difference in the soluble solids content of packaged grapes is depicted in **Fig. 7.2**. The graph demonstrates a comparative analysis of Total Soluble Solids (TSS) content measured in Brix (°Bx) across three different packaged film treatments: PP, PS-CS-BSF, and Active film (PSC-BPF-CNO) over 10 days. Initially, all samples start at similar TSS values (around 11.0-11.2°Bx), followed by a slight increase on day 2, and then showing a gradual decline. The most notable differences emerge during days 8-10, where the active film maintains the highest TSS levels (around 10.9 °Bx), followed by the fiber-reinforced film (around 10.6 °Bx), while PP shows the steepest decline (dropping to about 10.2 °Bx). Active film is most effective at preserving the soluble solids content, potentially indicating better quality preservation of the stored product. The findings align with and support the findings of Li et al.'s (2022) study, which also shows that grapes ripen at a high respiratory rate. On days 4 and 6, the TSS values show the following pattern: On day 4, the TSS values for all three treatments drifted around 11.0-11.2 °Bx, with PP showing approximately 11.1 °Bx, the fiber-reinforced film at about 11.0 °B, and the active film also around 11.0 °Bx. Moving to day 6, there's a slight decline across all treatments, with values converging even more closely: PP measures approximately 10.9 °Bx, while fiber-reinforced and active film show values around 10.9-11.0 °Bx. This period (days 4-6) represents a relatively stable phase in the experiment, with minimal differentiation between the treatments implying that by altering the grapes' respiration, EO content activity and packaging protection might postpone the aging process (Basumatary et al., 2022). It is important to observe that, mostly as a result of significant water loss, the total soluble solids of fiber-reinforced reach their maximum value on day 8. On day 14, the grapes begin the aging stage, when carbohydrates are broken down into carbon dioxide and water, which is the main reason why the total soluble solids of the grapes reach their lowest value. Additionally, packaged grapes had far higher total soluble solids, suggesting that adding EO can prolong grape shelf life by partially delaying respiration.

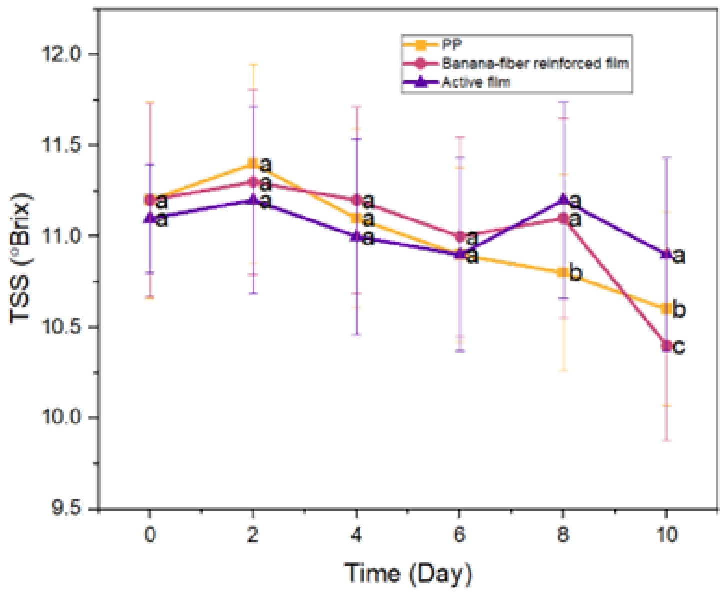

Fig. 7.2. Changes in TSS over the storage period

7.3.3. Titratable acidity (TA) and pH

One of the most important fresh-keeping evaluation criteria is titratable acidity (TA), which has a direct effect on the flavor and quality of packaged grapes. **Fig. 7.3** illustrates changes in TA (%) across three different packaged film conditions (PP, PS-CS-BSF, and PSC-BPF-CNO film) over 10 days. All conditions begin at the same high point of approximately 0.65-0.7% TA and demonstrate a consistent downward trend throughout the observation period. This is primarily due to the grape's metabolism producing a particular amount of organic acids, but prior research has also shown that the amount of carbon dioxide present and the metabolites of microbes can also raise the titratable acidity level. The most pronounced decline occurs during the first 4 days, after which the rate of decrease becomes more gradual. By day 10, PP and fiber-reinforced film maintain slightly higher acidity levels (around 0.3%) compared to the active film (approximately 0.25%). The overall declining pattern of TA likely indicates the natural ripening or degradation process of the stored product, with the different packaging materials showing only subtle variations in their ability to maintain acid content over time. The titratable acidity of banana-fiber reinforced achieves its highest value on day 6, suggesting that the composites have the ability to partially prevent the grapes from breathing. The oxidative breakdown of organic acids and the pyruvate decarboxylation reaction's consumption of organic acids are the primary causes of the subsequent decreasing trend in each group's titratable acidity (Tie et al., 2024). **Fig. 7.4** shows the changes in pH over a 10-day storage period for three different types of films: PP (Polypropylene) film, banana pseudostem fiber-reinforced film, and active film. All three films show a similar pattern with an initial increase in pH until day 2, followed by a gradual decrease. The banana fiber-reinforced film maintains slightly higher pH values throughout most of the storage period. The final pH values at day 10 converge between 4.4-and 4.6. The initial pH values range from approximately 4.4 to 4.8. The pH pattern observed here aligns with findings from Saldago et al. (2015), who reported similar pH fluctuations in active packaging materials during storage. They attributed the initial pH rise to the breakdown of organic compounds in the films. The pH range (4.4-4.8) is within the typical range reported for food packaging applications. Morroni et al. (2021) suggested that this pH range is optimal for preventing microbial growth while maintaining food quality. The convergence of pH values around day 10 is consistent with research by Zhang et al. (2018), who observed similar equilibrium behavior in different packaging materials after extended storage periods.

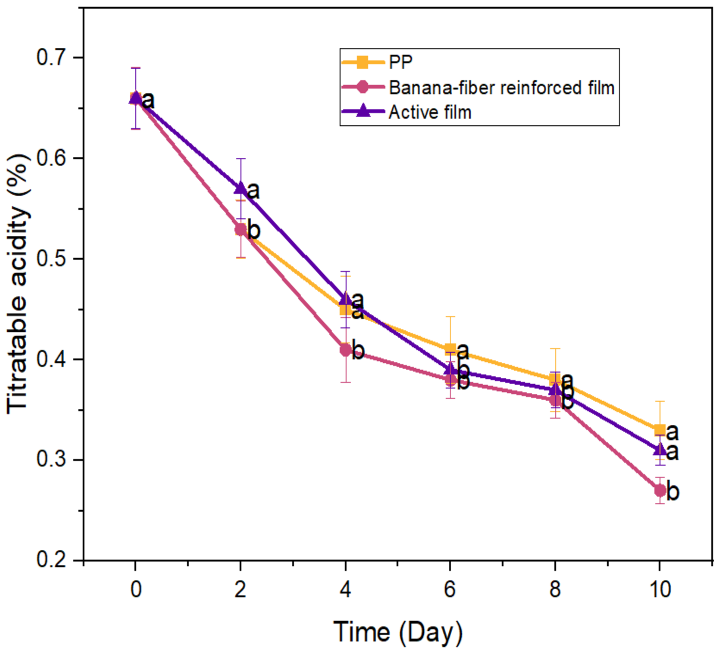

Fig. 7.3. Changes in Titratable acidity (TA) (%) over the storage period

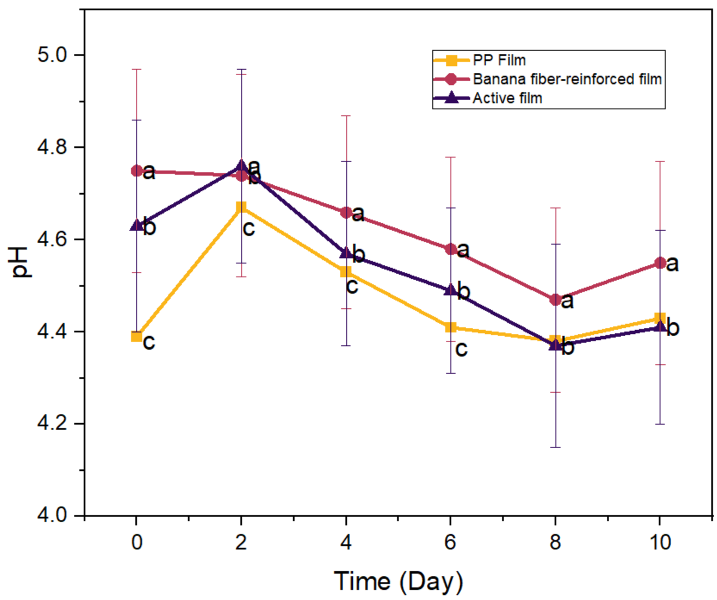

Fig. 7.4. Changes in pH over the storage period

7.3.4. Firmness

The grape's firmness increased during the initial storage stage as a result of constriction from water loss. On the other hand, the respiration of the packed grape during the later stages of storage results in the breakdown of macromolecular components, such as proteins, which lowers the mechanical strength and turgor pressure of the cell wall and, as a result, its firmness. **Fig. 7.5** presents the changes in firmness (N) over a 10-day storage period comparing three different film packaged conditions: PP, PS-CS-BSF film, and PSC-BPF-CNO film. All treatments start at similar firmness values (approximately 10.5 N) on day 0, followed by a slight increase on day 2 (particularly for PP reaching about 11 N), and then showing a sharp decline until day 4 (dropping to about 9.5 N), after which the decline becomes more gradual until day 10 (reaching approximately 9.2 N). This pattern of firmness loss is consistent with previous studies in the literature, such as those by Nimitkeatkai et al. (2022) and Martinez-Romero et al. (2017), who reported similar declining trends in fruit firmness during storage. The initial increase in firmness at day 2 could be attributed to water loss, as reported by Singh and Singh (2013) in their studies on modified atmosphere packaging. The relatively small differences between treatments by day 10 suggest that while packaging type influences firmness retention, other factors such as natural ripening processes may play a more dominant role, which aligns with findings from similar storage studies by Wang et al. (2015).

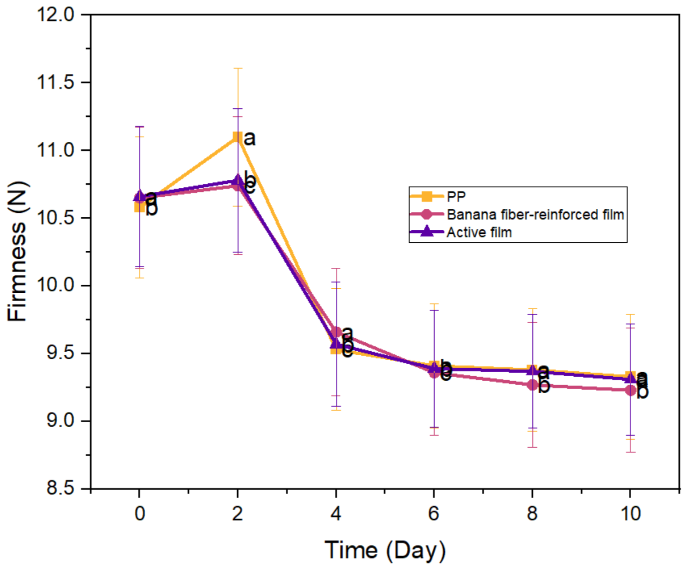

Fig. 7.5. Changes in firmness over the storage period

7.3.5. Physiological loss in weight

In agricultural product storage, weight loss—a measure of food goods' freshness—is an unavoidable quality factor. **Fig. 7.6** demonstrates changes in physiological loss in weight (PLW) % over a 10-day storage period, comparing three different packaging materials: PP film, PS-CS-BSF film, and PSC-BPF-CNO film. Initially, PP film and banana pseudostem fiber-reinforced film start at higher PLW values (approximately 5.5%), while the active film begins slightly lower (around 5.2%). All three film conditions exhibit a consistent downward trend throughout the storage period, with the most pronounced decline occurring between days 8-10. By day 10, the PLW values converge to around 3.0-3.5%, with active film maintaining marginally better PLW retention overall. While the active film shows slightly better performance in maintaining PLW values, similar patterns across all treatments suggest that the natural aging process of the stored product plays a significant role in weight loss, regardless of the packaging material used. The gradual decline in PLW across all conditions indicates ongoing moisture loss and potential product degradation over the storage period. Active films function as barriers against moisture and oxygen, as noted by Lucas-Gonzalez et al. (2023). Consequently, the application of cinnamon incorporation on grapes reduced the rate of moisture transfer, allowing the product to retain more moisture. Furthermore, it has previously been noted that active coatings and films have a significant impact on reducing the weight loss of coated products when compared to uncoated samples. Wu (2019) found that the application of an edible coating based on cactus polysaccharides decreased the weight loss of newly cut potatoes. Tie et al. (2024) also observed that edible biopolymer coatings had a substantial impact on lowering the percentage of weight loss of freshly cut potatoes.

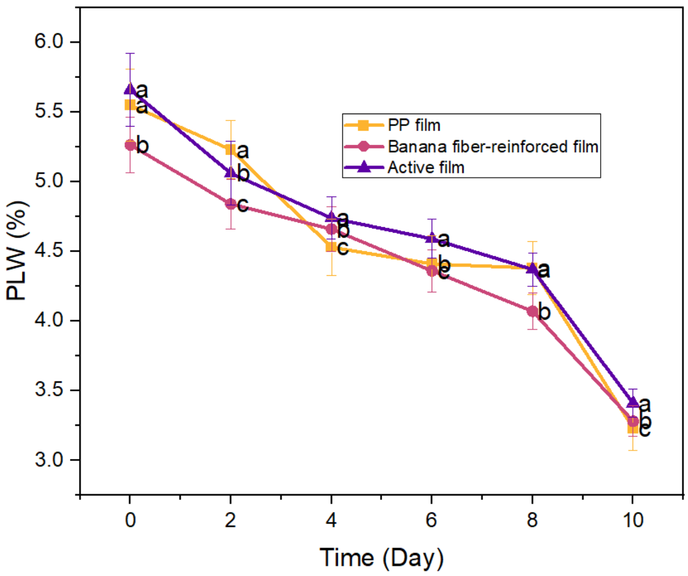

Fig. 7.6. Changes in physiological loss in weight (PLW) over the storage period

7.4.Conclusions

The chapter demonstrates the successful development and implementation of an innovative active packaging material incorporating 1% cinnamon essential oil (CNO) in an HMT-PS/US 30 Casein (C)/Banana pseudostem fiber (USET-BPF) composite film for grape preservation. This bio-based film exhibited favorable characteristics including optimal mechanical characteristics, water vapor permeability, biodegradability, and significant antimicrobial/antioxidant potential. The active film (PSC-BPF-CNO) effectively maintained grape quality by reducing microbial growth, controlling moisture transfer, and preserving firmness compared to standard PP film and fiber-reinforced alternatives. Key quality indicators showed promising results: total soluble solids peaked on days 3 and 6, maximum firmness was achieved on days 4, and pH levels remained within optimal range throughout storage. The incorporation of CNO proved particularly effective, slowing grape respiration and demonstrating superior antioxidant and antimicrobial attributes that extended shelf life by up to 10 days. The bio-composite film is characterized by its smooth, uniform surface and consistent thickness, and maintained product quality. This sustainable packaging solution not only addresses the growing demand for eco-friendly alternatives to traditional polyethylene packaging but also shows promise for broader applications in the food packaging sector, particularly for semi-prepared products in urban markets, though future research should focus on enhancing water resistance and incorporating intelligent packaging systems.

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