

# Fruit Preservative Composed of Bamboo Vinegar, Polyvinyl Alcohol, and Citric Acid for Mulberry Preservation

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In order to solve the problem of spoilage of mulberry after harvest, this study discusses the effect of various concentrations of composite preservatives on the preservation effect of mulberry, so as to achieve the purpose of prolonging its shelf life. Taking mulberry as raw material, three preservatives of bamboo vinegar, polyvinyl alcohol, and citric acid were selected, and the physical and chemical indexes used were sensory evaluation, firmness, weight loss rate, decay percentage, soluble solid content, vitamin C content, and POD activity, and the optimal concentration of each preservative was determined to be 1.0% bamboo vinegar, 3.0% polyvinyl alcohol (PVOH), and 1.0 % citric acid by single factor test at room temperature. The orthogonal test screened 1.0% bamboo vinegar, 3.5% PVOH, and 0.8% citric acid as the optimal compound combination, which could effectively extend the shelf life of mulberry for at least 48 h compared to the blank control and commercial groups. This study provided a safe and efficient compound preservative solution for mulberry preservation.

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**Keywords:** Bamboo vinegar; Polyvinyl alcohol; Citric acid; Mulberry preservation

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## INTRODUCTION

Mulberry fruit, the mature aggregate fruit of *Morus alba* L. from the Moraceae family, is rich in various active nutritional components and possesses both edible and medicinal value. Mulberries are typical respiratory leap fruits. During the post-harvest ripening process, there will be a distinct respiratory peak, with a sharp increase in ethylene release (Hao *et al.* 2022). Its thin and tender peel, coupled with fragile tissue, makes it highly susceptible to mechanical damage, water loss and wrinkling, softening and decay, as well as rapid deterioration of nutritional quality during postharvest handling. At ambient temperature, the shelf life of mulberries is typically only 1 to 2 days, which severely limits their commercial distribution and market value (Qiao *et al.* 2021). Therefore, developing efficient, safe, and economical preservation technologies is of significant importance for the sustainable development of mulberry industry.

Currently, mulberry preservation primarily employs chemical, physical, and biological methods, yet each has certain limitations. Although chemical preservation is operationally convenient and effective, its long-term use may pose risks of chemical residues and environmental pollution (Liu 2025). For instance, treatment with specific concentrations of 1-hexanol can effectively suppress the respiratory intensity of mulberries, and slow down the degradation of substances such as vitamin C (Huo *et al.* 2012). Controlled atmosphere storage can be carried out by regulating oxygen and carbon dioxide concentrations, thereby creating a microenvironment that inhibits respiration. Research has confirmed its efficacy in maintaining fruit firmness and antioxidant capacity (Abey Suriya *et al.* 2024). Among biological methods, edible coatings, such as nanocomposite films of cellulose nanofibrils and chitosan, can effectively reduce weight loss and decay rate during mulberry storage (Shang 2022). However, achieving a balance between their mechanical and barrier properties remains a challenge. Against this backdrop, natural extracts have become a research focus due to their green and safe characteristics. Bamboo vinegar as a natural liquid by-product produced by the high-temperature pyrolysis of bamboo materials. Its main components are water and organic acids.

The three materials (bamboo vinegar, polyvinyl alcohol (PVOH), and citric acid) have a wide range of sources, and each has distinct characteristics. Bamboo vinegar is a complex dark-brown liquid primarily composed of over 200 compounds, including organic acids (mainly acetic acid), phenols, and alcohols (Qian *et al.* 2012). The core of its preservative action lies in dual mechanisms of antibacterial and antioxidant activities. Studies have shown that bamboo vinegar can effectively scavenge superoxide anion radicals, hydroxyl radicals, and DPPH radicals, while also demonstrating reducing power towards potassium ferricyanide (Li *et al.* 2023). Together, these effects provide a theoretical basis for delaying oxidative deterioration in fruits. Regarding its antibacterial properties, its organic acid components can dissociate hydrogen ions, lower the intracellular pH of microorganisms, and disrupt their enzymatic systems. Simultaneously, its abundant phenolic substances can interfere with the integrity of microbial cell membranes, increasing their permeability and ultimately leading to the leakage of cellular contents. PVOH is a water-soluble synthetic polymer known for its excellent film-forming ability, biocompatibility, and chemical stability (Huang *et al.* 2024; Li *et al.* 2024). Its core function within the preservation system is physical barrier formation: it can create a transparent, flexible, and dense continuous film on the fruit surface. This film effectively blocks the diffusion of external oxygen into the fruit. It simultaneously reduces the evaporation and loss of internal moisture, thereby dually inhibiting the fruit's respiration and transpiration processes, thus delaying senescence. Citric acid, a natural organic acid, plays a key role of chemical regulation within the composite preservation system. It can lower the environmental pH, synergistically enhancing the antibacterial effect of other agents (Yu *et al.* 2023). More importantly, its strong metal ion chelating ability can effectively inhibit the activity of key enzymes such as polyphenol oxidase, thereby blocking the enzymatic browning reaction chain. In a study, researchers pointed out that citric acid treatment effectively delayed fruit deterioration and inhibited the occurrence of superficial browning and decay (Nongnual *et al.* 2024). However, there have been no reports on the combinations of these three materials in the preservation and freshness-retention fields.

Based on the functional complementarity of the aforementioned three materials, this study innovatively combined them to construct a "physical-chemical-biological"

ternary synergistic preservation system. This system employed a multi-layer protection design: the outer layer consists of the PVOH physical barrier film; the middle layer is a functional sustained-release layer comprising bamboo vinegar and citric acid; the inner layer relies on the penetration of citric acid to inhibit enzymatic browning. The research would optimize the formulation ratio through single-factor and orthogonal experiments, and evaluate the preservation effect using key indicators such as decay percentage, weight loss rate, firmness, soluble solid content, vitamin C content, and peroxidase (POD) activity. This study aimed to develop a safe, efficient, and economical mulberry-specific preservative, while also providing a new pathway for the high-value utilization of bamboo vinegar.

## EXPERIMENTAL

### Materials

Bamboo vinegar (mass concentration: 5.0%, organic acid $\geq$ 4.0%) was provided by Suichang Wutailang Bamboo Charcoal Co., Ltd. (Zhejiang, China). Polyvinyl alcohol and citric acid were purchased from Shanghai Macklin Biochemical Technology Co., Ltd. (Shanghai, China). Oxalic acid was supplied by Tianjin Baisht Chemical Co., Ltd. (Tianjin, China). 2,6-Dichloroindophenol sodium salt was sourced from Feiyang Biotechnology Co., Ltd. (Guangzhou, China). Vitamin C was obtained from Cool Chemical Technology Co., Ltd. (Beijing, China). Fresh mulberry fruits (diameter: 1.5-2.0 cm) for the experiments were supplied by Lanxi Xingyao Trade Co., Ltd. (China). The commercially available preservative (Fruit and vegetable preservative, main ingredient: antioxidants, fungicides) was provided by Ruzhou Huaduoduo Biotechnology Co., Ltd. (Henan, China). The water used throughout the experiments was distilled water at room temperature.

All the experiments below were conducted in triplicate.

### Single-Factor Experiment

Mulberry fruits of uniform maturity and size, free from mechanical damage, were selected. They were then cleaned and air-dried naturally in a well-ventilated area. The mulberries were immersed in each preservation solution for 5 min, with immersion in clean water serving as the control. After removal from solution, mulberries were air-dried in a ventilated area to remove surface moisture, then packaged separately in food-grade polypropylene (PP) containers and stored at room temperature (20 to 30 °C) for 7 d. Sensory evaluation and determination of weight loss rate were conducted at room temperature by 24 h intervals, and the average values were calculated to screen for the optimal concentration of each preservative.

Preservation solutions were prepared with different concentrations, as follows: bamboo vinegar: 0.5%, 1.0%, 1.5%; PVOH: 1.0%, 2.0%, 3.0%; Citric acid: 0.5%, 1.0%, 1.5%. The PVOH solutions were heated in a water bath at 90 °C and stirred with 200 rpm until no obvious particles were visible.

### Orthogonal Experiment

Based on the results of the single-factor experiments, gradients of  $\pm 0.2\%$  for bamboo vinegar and citric acid, and  $\pm 0.5\%$  for PVOH were established. The optimal concentrations of each preservative are presented in Table 1. The composite preservatives

was obtained as follows: PVOH solutions were heated in a water bath at 93 °C and stirred with 300 rpm, then bamboo vinegar and citric acid was added into the solution with stirring state, respectively. Nine composite preservative formulations were designed using an L9 (3<sup>4</sup>) orthogonal array (Table 2). Simultaneously, a clean water control group and a commercially available preservative group (prepared by diluting with water according to the instructions) were set up. Mulberries from each group were stored at room temperature (20 to 30 °C) for 7 d. Sensory scores, firmness, weight loss rate, decay percentage, vitamin C content, and POD activity were measured every 24 h, and the average values were calculated. The optimal composite preservative ratio was determined through data analysis.

**Table 1.** Optimal Concentrations of Each Preservative

|                       | Bamboo Vinegar (%) | Polyvinyl Alcohol (%) | Citric Acid (%) |
|-----------------------|--------------------|-----------------------|-----------------|
| Optimal concentration | 1.0                | 3.0                   | 1.0             |

**Table 2.** Orthogonal Experiment Table of Composite Preservatives on Mulberry Preservation

| Experimental Group Number | Bamboo Vinegar (%)            | Polyvinyl Alcohol (%) | Citric Acid (%) |
|---------------------------|-------------------------------|-----------------------|-----------------|
| 1                         | 0.8                           | 2.5                   | 0.8             |
| 2                         | 0.8                           | 3.0                   | 1.0             |
| 3                         | 0.8                           | 3.5                   | 1.2             |
| 4                         | 1.0                           | 2.5                   | 1.0             |
| 5                         | 1.0                           | 3.0                   | 1.2             |
| 6                         | 1.0                           | 3.5                   | 0.8             |
| 7                         | 1.2                           | 2.5                   | 1.2             |
| 8                         | 1.2                           | 3.0                   | 0.8             |
| 9                         | 1.2                           | 3.5                   | 1.0             |
| 10                        | Blank control group           |                       |                 |
| 11                        | Commercial preservative group |                       |                 |

### Sensory Evaluation

Ten panelists were randomly invited to assign sensory scores according to Table 3, and the average value was calculated as the sensory score for each sample.

**Table 3.** Mulberry Sensory Evaluation Score Table

| Evaluation Indicators | Standard                                                | Scores       |
|-----------------------|---------------------------------------------------------|--------------|
| Appearance            | Dark purplish-red, Uniform and without brown spots      | 20-30 points |
|                       | slight browning                                         | 10-20 points |
|                       | Widespread browning                                     | 1-10 points  |
| Odor                  | Fresh and clean mulberry aroma, with no off-odor        | 20-30 points |
|                       | Weakened aroma, with a slight grassy note               | 10-20 points |
|                       | Pronounced rancid or moldy odor                         | 1-10 points  |
| Texture               | Plump, intact, glossy, no visible droplets in container | 20-30 points |
|                       | Slightly dull or shriveled, few droplets in container   | 10-20 points |
|                       | Severely shriveled, with heavy container condensation   | 1-10 points  |
| Taste                 | Sweet and juicy                                         | 20-30 points |
|                       | Good taste                                              | 10-20 points |
|                       | Taste was poor                                          | 1-10 points  |

### Fruit Firmness

Firmness was measured using a pointer-type hardness tester (GY-3, Yueqing Aidebao Instrument Co., Ltd, Zhejiang, China). The probe was pressed vertically and uniformly into the mulberry surface until the scale line was reached, and the firmness value (kg/cm<sup>2</sup>) was recorded. The measurement was repeated three times for each group, and the average value was calculated.

### Fruit Weight Loss

The weight loss of mulberries was determined gravimetrically. Each empty container was weighed, then weighed again with the mulberries. The weight loss was calculated as a percentage of the initial weight, and the average for each group was recorded.

### Fruit Decay Percentage

The decay percentage was determined according to the method described by Tu *et al.* (2024), with decay being recorded when the mildewed and rotten area on the mulberry surface exceeded one-quarter of the total surface area.

### Vitamin C Content Determination

10 g of destemmed mulberry pulp was weighed, and a small amount of oxalic acid solution was added. Then the mixture was ground into a pulp. The mulberry pulp was poured into a 100 mL volumetric flask and diluted it to 100 mL with oxalic acid solution. 10 mL of the diluted mulberry-like liquid was poured into a 50 mL conical flask, and titrated with sodium 2, 6-dichlorophenol solution. When the solution color began to turn pink and the pink color did not fade after shaking the conical flask for 15 s, the titration was completed and the data was recorded. The Vc content was calculated as formula (1),

$$V_c(\text{mg}/100\text{g}) = \frac{(V - V_0) * T * A}{W} \times 100 \quad (1)$$

where  $V$  is the volume of sodium 2,6-dichlorophenol solution consumed when titrating mulberry solution (mL),  $V_0$  is the volume of sodium 2,6-dichlorophenol solution consumed in the titration blank test (mL),  $T$  is titration degree (mg/mL),  $A$  is dilution ratio of mulberry juice, and  $W$  was the weight of mulberry (g).

### Peroxidase Activity

The POD activity was determined as previously described (Zhang *et al.* 2022; Lü *et al.* 2024), with minor modifications. A 0.5 g sample of mulberry fruit was taken and cut into pieces. It was then homogenized with 6 mL of pre-cooled 0.1 mol/L phosphate buffer (pH=7.0) in an ice bath. The homogenate was filtered through four layers of gauze and centrifuged at 4000 r/min for 15 min. The supernatant was collected as the crude enzyme extract; it was diluted appropriately if the color was too deep. For the assay, 5 mL of the reaction mixture, consisting of 3 mL of phosphate buffer, 1 mL of 0.05 mol/L guaiacol, and 1 mL of 2% hydrogen peroxide, was thoroughly mixed. Then, 1 mL of the crude enzyme extract was added and mixed immediately. The absorbance was measured at 470 nm, with readings recorded every 30 s for a total of 3 min. A blank control, using buffer instead of the enzyme extract, was used to zero the spectrophotometer. One unit of enzyme activity (U) was defined as the amount of enzyme causing a change of 0.01 in

absorbance per minute, and the result was expressed as  $\text{U} \cdot \text{g}^{-1} \cdot \text{min}^{-1}$ . The POD activity was calculated as formula (2):

$$\text{POD activity} = \frac{\Delta A_{470} \times V_T}{m \times V_s \times 0.01 \times t} \quad (2)$$

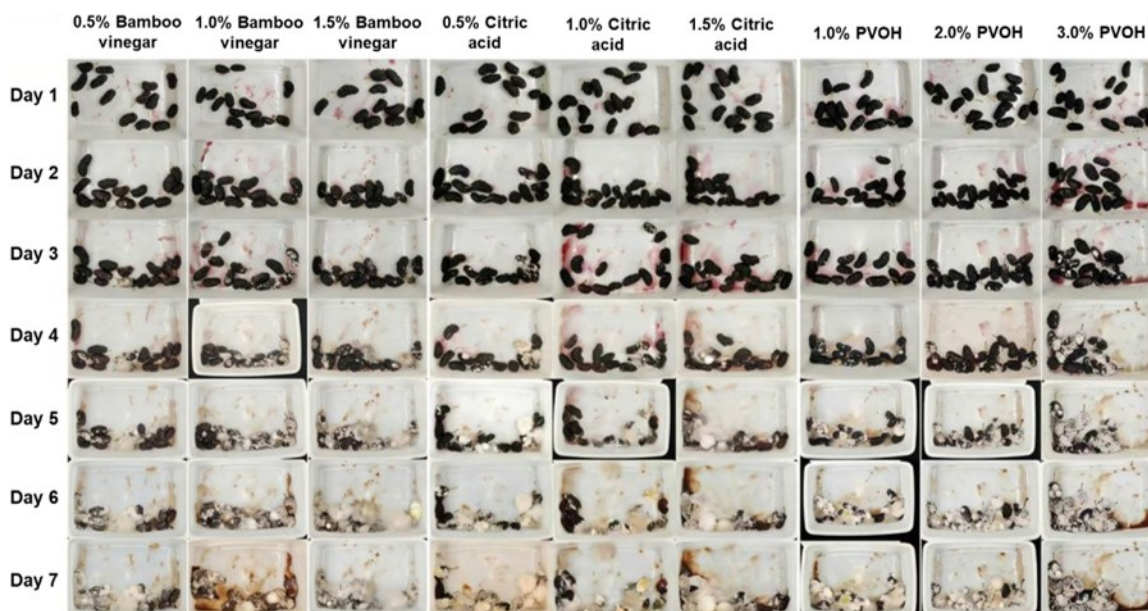
In Eq. 2,  $\Delta A_{470}$  is the change in absorbance during the reaction time,  $V_T$  is the total volume of extracted mulberry juice (mL),  $m$  is quantity of mulberry (g),  $V_s$  is the volume of crude enzyme liquid used in the determination (mL), and  $t$  is reaction time (min).

## RESULTS AND DISCUSSION

### Analysis of Single-Factor Experiments

#### Sensory effects

Sensory quality, encompassing appearance, texture, and taste, is a critical factor influencing consumers' desire to purchase fruit (Wang *et al.* 2025). To investigate the preservative effect of different agents on mulberries, this study conducted sensory evaluation focusing mainly on their appearance and texture. As shown in Fig. 1, after treatment with different concentrations of bamboo vinegar, all mulberries remained in good condition for the first two days, with browning and mycelium appearing on the third day. Plump, intact, glossy, no visible droplets in container were observed on the second day, while slightly dulled or shriveled, and few droplets appeared in the container from the third day.



**Fig. 1.** Morphological changes of mulberries treated with different concentrations of bamboo vinegar, citric acid, and polyvinyl alcohol preservation solutions during storage

The decay percentage of the 1.0% concentration group was lower than that of the 0.5% and 1.5% groups. The 0.5% group exhibited browning due to its lower concentration and insufficient antioxidant components; the 1.5% group, however, may

have experienced accelerated water loss because its excessively high acidic components and phenolic compounds potentially damaged the epidermal cells. Therefore, 1.0% bamboo vinegar demonstrated the optimal preservative effect. In the PVOH treatments, all mulberry groups performed well on the first day, with differences emerging on the third day. The 1.0% and 2.0% groups, which formed thinner films with insufficient barrier properties, showed higher rates of mold growth and decay compared to the 3.0% group, indicating that 3.0% PVOH provided the best preservation. For citric acid treatments, all mulberries displayed bright color on the first day. On the second day, the 0.5% group, due to its low concentration, was the first to develop mycelium. The 1.0% and 1.5% groups only began to show sporadic mycelium on the third day, suggesting that the antimicrobial effect of higher citric acid concentrations diminished over time. By the seventh day, mulberries in all groups were largely completely decayed; Overall, the preservative effect of the 1.0% citric acid group was superior to that of the other two groups.

#### Fruit weight loss

Weight loss is a critical indicator for assessing the quality loss of mulberries during storage caused by factors such as water evaporation and respiratory consumption. Due to the thin and delicate peel of mulberries, investigating changes in their weight loss is of great significance for preservation (Luo *et al.* 2022). As shown in Fig. 2, on day 7, the 1.0% PVOH group exhibited the highest weight loss. This might be because the concentration was too low to form a continuous and dense coating film on the mulberry surface, resulting in a porous membrane structure with cracks that could not effectively barrier against water evaporation. For the three bamboo vinegar groups, the weight loss values from high to low were 1.5%, 0.5%, and 1.0%; for the polyvinyl alcohol groups, they were 1.0%, 2.0%, and 3.0%; and for the citric acid groups, they were 1.5%, 0.5%, and 1.0%. A high weight loss rate indicates significant water evaporation, making the fruit prone to shriveling and deterioration, thus reflecting poor preservation efficacy. Consequently, 1.0% bamboo vinegar, 1.0% citric acid, and 3.0% PVOH demonstrated better preservation effects on mulberries.

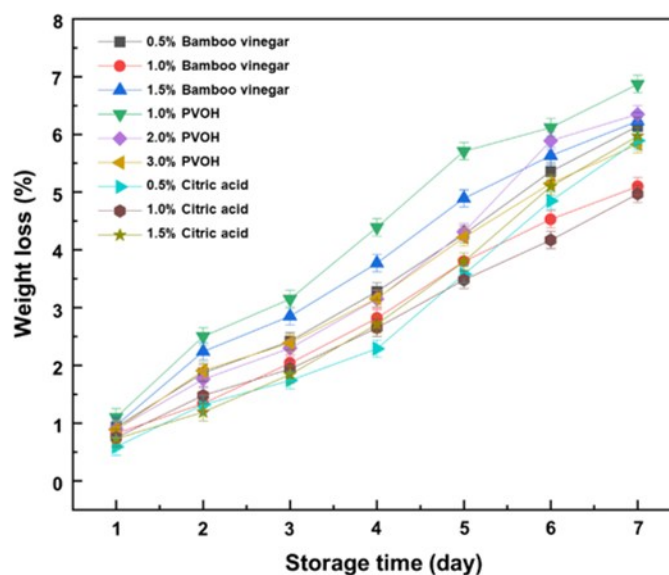


Fig. 2. Single-factor experiment on the weight loss of mulberry during storage

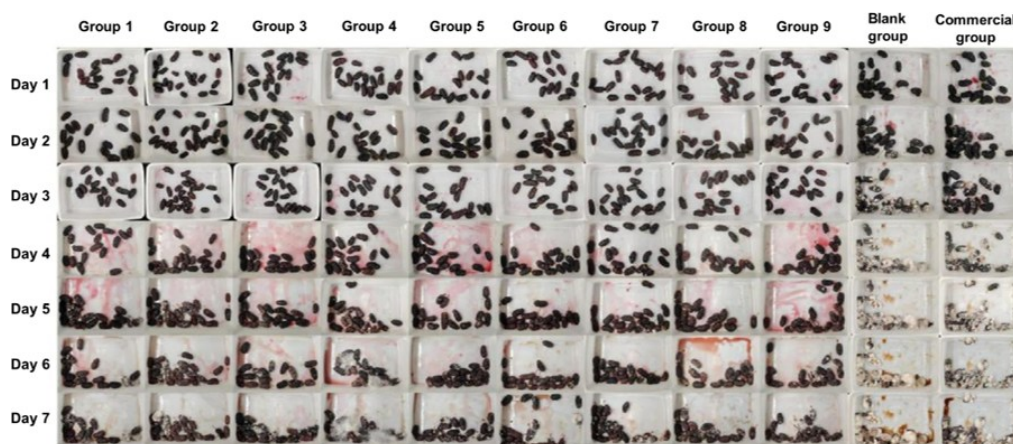
### Comprehensive analysis of single-factor experiments

Based on a comprehensive analysis of the sensory evaluation and weight loss indicators from the single-factor experiments, the optimal concentrations of bamboo vinegar, citric acid, and PVOH were determined to be 1.0%, 1.0%, and 3.0%, respectively. Using these as a foundation, the concentration gradients were refined to design the orthogonal experiment.

## Analysis of Orthogonal Experiment

### Sensory effects

As shown in Fig. 3, significant differences in morphology were observed between the treatment groups and the control group during storage. From day 1 to 3, the mulberries in all composite preservative treatment groups maintained an intact and plump surface, with no signs of juice exudation or browning. This indicated that the physical barrier formed by PVOH effectively delayed water evaporation and oxygen penetration, while the antimicrobial components of bamboo vinegar inhibited early microbial colonization. Starting from day 4, the treatment groups began to show slight juice exudation and a mild sour odor. Among these, groups 2, 5, and 6 exhibited sporadic mycelial growth on the surface; however, the overall decay percentage remained significantly lower than that of the control group. By day 7, mulberries in the treatment groups showed partial decay, primarily concentrated around the fruit stem or at sites of mechanical damage, and the decay process was slow. This suggested that the composite preservative effectively inhibited microbial proliferation and enzymatic browning. In contrast, the commercial preservative group and the blank control group displayed juice exudation and localized decay as early as day 2, with mycelium spreading rapidly. By day 4, the blank control group was almost completely decayed. In summary, the composite preservative, through its synergistic mechanism of “physical barrier (PVOH) + chemical antimicrobial action (bamboo vinegar) + antioxidant regulation (citric acid),” significantly inhibited the decay process of mulberries.



**Fig. 3.** The morphological changes of mulberries after treatment with preservation solution in experimental groups 1-9, blank group, and commercial group during storage. Group 1: 0.8% bamboo vinegar, 2.5% PVOH, 0.8 % citric acid; Group 2: 0.8% bamboo vinegar, 3.0% PVOH, 1.0 % citric acid; Group 3: 0.8% bamboo vinegar, 3.5% PVOH, 1.2 % citric acid; Group 4: 1.0% bamboo vinegar, 2.5% PVOH, 1.0 % citric acid; Group 5: 1.0% bamboo vinegar, 3.0% PVOH, 1.2 % citric acid; Group 6: 1.0% bamboo vinegar, 3.5% PVOH, 0.8 % citric acid; Group 7: 1.2% bamboo vinegar, 2.5% PVOH, 1.2 % citric acid; Group 8: 1.2% bamboo vinegar, 3.0% PVOH, 0.8 % citric acid; Group 9: 1.2% bamboo vinegar, 3.5% PVOH, 1.0 % citric acid

### Sensory scores

As shown in Fig. 4, on the 7<sup>th</sup> day of storage, Group 6 exhibited the highest sensory score, followed by Group 9, while Group 10 and Group 11 had the lowest scores, indicating that the preservative prepared in this study had better preservation effect. All 9 experimental groups scored higher than the control groups. Both Group 6 and Group 9 utilized 3.5% PVOH, whereas Group 4, which had the lowest score, used a PVOH concentration of 2.5%. This indicated that a higher concentration of PVOH facilitated the formation of a dense film layer, effectively delaying water evaporation and oxygen penetration, thereby mitigating browning and softening. This finding was consistent with the research conclusions of Liu *et al.* (2025b). The relatively low score of the commercial preservative may be attributed to its general-purpose formulation, which was poorly adapted to the high-moisture and thin-skin characteristics of mulberries, resulting in inferior barrier properties and antimicrobial effects.

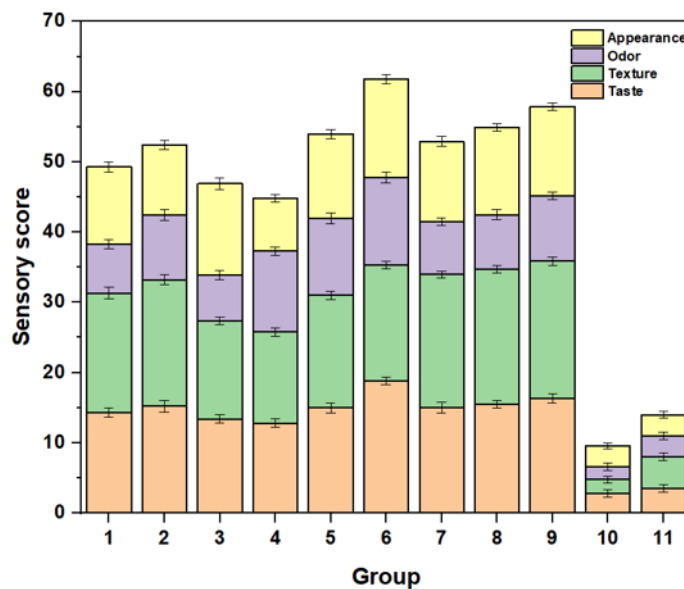
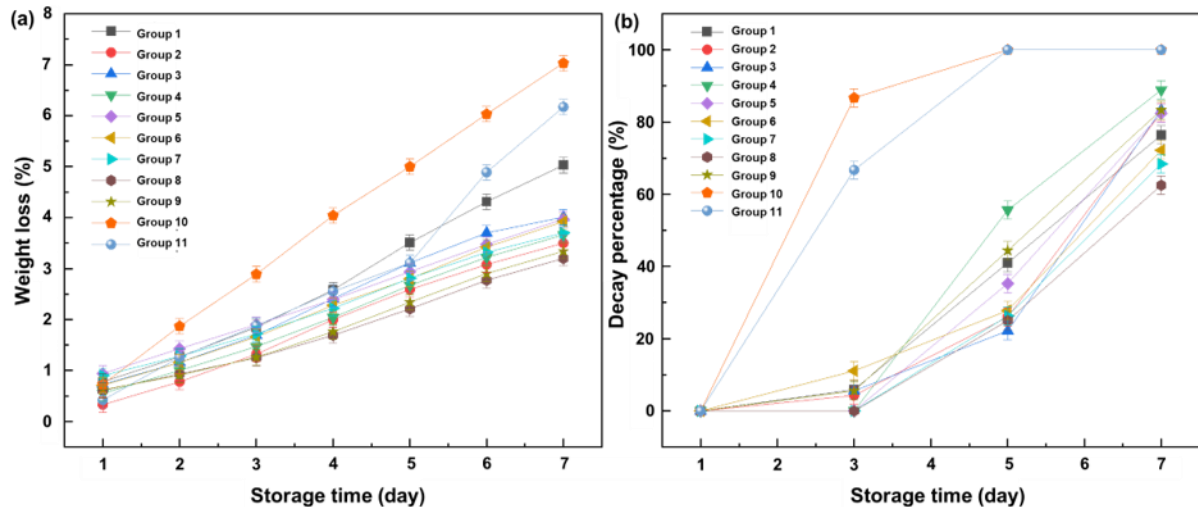


Fig. 4. Mulberry sensory score of mulberry on the 7-day storage

### Fruit weight loss rate and decay percentage

As shown in Fig. 5a, b, the weight loss rate of mulberries in all groups except Group 11 increased with storage time, exhibiting an approximately linear trend. The blank control group had the highest weight loss rate, followed by the commercial preservative group. The nine composite preservative groups showed relatively small differences, with Group 8 exhibiting the lowest weight loss rate. This phenomenon may be attributed to the thin peel of mulberries and their inherently weak resistance to water loss. Regarding the decay percentage, all composite preservative groups demonstrated significantly lower decay percentage compared to the commercial and blank control groups, with particularly pronounced inhibitory effects during the early storage period. On day 3, Group 6 had the highest decay percentage among the experimental groups, was approximately 15%, whereas the commercial and blank control groups reached 65% and 70%, respectively. By day 7, Group 8 exhibited the lowest decay percentage, extending the shelf life by at least 48 h compared to the blank control and commercial groups. Although the commercial group performed better than the blank control group during the first 4 days, it decayed rapidly after day 5 due to its single antimicrobial mechanism lacking a physical barrier. In contrast, the experimental groups effectively delayed

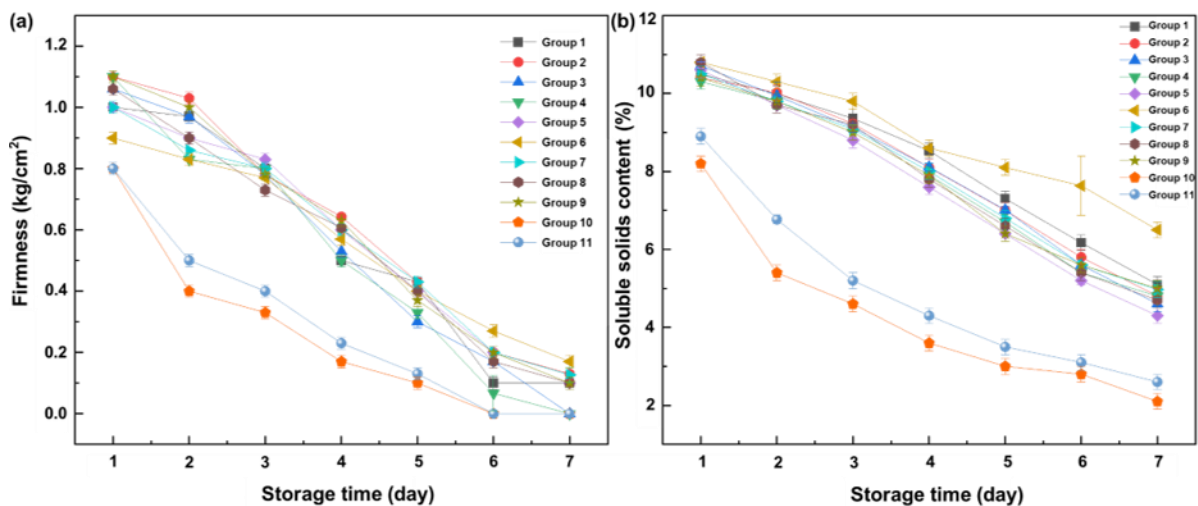
spoilage through the synergistic effects of the physical barrier provided by PVOH, the antimicrobial action of bamboo vinegar, and the antioxidant regulation of citric acid. Among these, Group 8 formulated with an optimal ratio including 3.5% PVOH, demonstrated the best performance.



**Fig. 5.** Orthogonal experiment on the (a) weight loss, (b) decay percentage of mulberry during storage

#### *Firmness and soluble solids content*

Mulberry is a highly perishable fruit. During its storage, firmness serves as a critical indicator of quality deterioration (Liu *et al.* 2025a). The experimental data showed (Fig. 6a) that the firmness of the compound preservative-treated groups remained significantly higher than that of the commercial and blank control groups throughout the storage period, with minimal differences observed among the various treatment groups. This indicated that the compound preservative effectively maintained the structural integrity of fruit cells through synergistic multi-mechanisms.

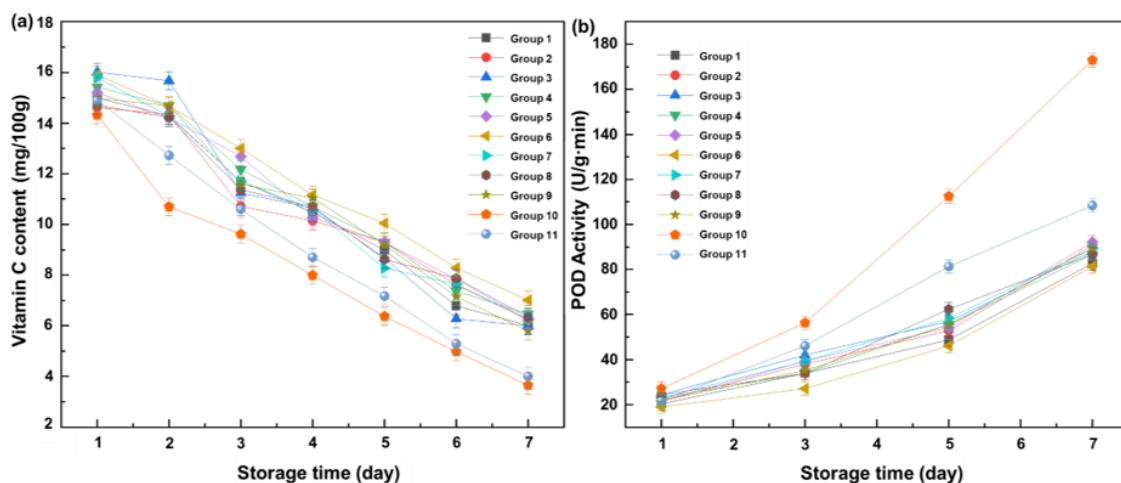


**Fig. 6.** (a) Orthogonal experiment on the (a) firmness, (b) soluble solids content of mulberry during storage

Group 6 exhibited the highest firmness. In contrast, firmness in group 10 and group 11 declined to zero by day 6. The commercial group, lacking physical barriers and antioxidant components, relied solely on chemical preservatives for short-term antimicrobial effects and could not prevent water loss or cell wall degradation, whereas the blank group remained completely unprotected. Soluble solids content is an important parameter for evaluating fruit texture and flavor (Wang *et al.* 2025). As storage time progressed, its content gradually decreased due to respiratory consumption. On day 1, the content in the first nine groups ranged from 10% to 11%, while both control groups were below 9%. By day 7, group 6 maintained a significantly higher soluble solids content compared to the other groups (Fig. 6b). In conclusion, the compound preservative effectively delayed the quality deterioration of mulberries through synergistic effects, with group 6 demonstrating the best overall performance.

### Vitamin C Content and Peroxidase Activity

Vitamin C (Vc) is an important nutritional component in fruits, known for its antioxidant properties (Cheng *et al.* 2025). Experimental results showed (Fig. 7a) that the Vc retention rate in the composite preservative treatment groups during storage was significantly superior to that of the commercial and blank control groups, with Group 6 exhibiting the best effect. On day 1, the Vc content in all groups was similar (approximately 15 mg/100 g), subsequently decreasing gradually due to oxidation and enzymatic degradation. By day 7, the Vc content in Group 6 remained close to 7 mg/100 g, significantly higher than that of the other groups. POD activity is an oxidoreductase found in fruits and vegetables; an increase in its activity typically exacerbates browning and is negatively correlated with preservation efficacy (Jiang 2024). As shown in Fig. 7b, the POD activity in the nine experimental groups remained consistently lower than that of the control groups throughout storage, indicating that the preservative effectively inhibited the rise in POD activity. Among these, Group 6 maintained the lowest POD activity throughout the storage period, demonstrating the optimal preservation effect. In conclusion, the composite preservative delayed the senescence of mulberries by reducing Vc consumption and inhibiting POD activity, with Group 6 showing the best overall performance.



**Fig. 7.** (a) Orthogonal experiment on the vitamin C content of mulberry, (b) Orthogonal experiment on the POD activity of mulberry

## CONCLUSIONS

1. Bamboo vinegar and citric acid synergistically delayed the quality deterioration of mulberries through multiple mechanisms. The phenolic compounds in bamboo vinegar mitigated browning and spoilage by scavenging free radicals and inhibiting microbial proliferation. Citric acid suppressed enzyme activity by modulating the microenvironmental pH and chelated metal ions, thereby disrupting the oxidative reaction chain. The dense film layer formed by polyvinyl alcohol (PVOH) reduced water evaporation and oxygen permeation through physical barrier effects, maintaining cell turgor and structural integrity, and synergistically inhibited metabolic losses.
2. Individual preservative agents have inherent limitations, while their combined usage may significantly enhance overall preservation performance. Single-factor experiments indicated that the optimal concentrations were 1.0% for bamboo vinegar, 3.0% for PVOH, and 1.0% for citric acid. Through orthogonal experiment analysis of various indicators-including sensory score, weight loss rate, firmness, vitamin C content, and POD activity-the optimal composite formulation was determined to be 1.0% bamboo vinegar combined with 3.5% PVOH and 0.8% citric acid. This formulation achieves an optimal balance in mulberry preservation efficacy through a synergistic mechanism of “physical barrier, chemical antimicrobial action, and antioxidant regulation”.
3. The concentration of preservative agents requires scientific regulation, as both excessive and insufficient amounts can influence preservation efficacy. High concentrations of bamboo vinegar can easily lead to osmotic imbalance in the fruit peel, accelerating juice exudation. Excessive PVOH concentration may induce anaerobic respiration due to insufficient film permeability, leading to the accumulation of toxic metabolites such as ethanol, while concentrations that are too low fail to provide adequate barrier effects. Excessive citric acid concentration may disrupt cell membrane structure, exacerbating fruit softening. Therefore, achieving an optimal formulation ratio is critical to maximizing the effectiveness of preservative.

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