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Strategy to increase the fresh durability of water spinach (*Ipomea aquatica*) at a room temperature of 10° C using packaging technology

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Abstract

Water spinach (*Ipomoea aquatica*) is a leafy vegetable widely consumed in Southeast Asia; however, it rapidly deteriorates after harvest. This study evaluated the effect of passive modified atmosphere packaging (MAP) using low-density polyethylene (LDPE) films—both unperforated and perforated with 1% holes—on the postharvest quality of water spinach during cold storage at 10 ± 1 °C. The experiment was arranged in a completely randomized design with two packaging treatments and three sampling intervals (0, 5, and 10 days). The LDPE film without holes (T₁) created a passive modified atmosphere characterized by a gradual decrease in O₂ levels (to approximately 8%) and a concomitant increase in CO₂ levels (to approximately 3%), whereas the LDPE film with holes (T₂) maintained gas levels close to ambient. A significant interaction between treatment and time (p < 0.001) was observed for chlorophyll content, gas composition, and storage losses. Water spinach in T₁ exhibited lower cumulative storage losses and better chlorophyll retention compared to T₂, indicating a slower physiological decline under reduced O₂ and elevated CO₂ conditions. Overall, passive modified atmosphere packaging (MAP) using non-perforated LDPE film effectively maintained visual freshness and minimized water spinach quality losses for up to 10 days. Based on the comparison of T₁ and T₂ treatments, T₁ was superior, as observations up to the tenth day showed only a slight percentage loss in T₁ relative to T₂, likely due to the completely sealed packaging used in T₁.

Keywords: Atmosphere, Modification, Respiration, MAP, Low-Density Polyethylene

1. Introduction

Water spinach (*Ipomoea aquatica*) is one of the most popular leafy vegetables in Southeast Asia. This vegetable is valued for its tender leaves and is rich in chlorophyll, vitamins, and minerals, such as calcium (Putri et al., 2024). Water spinach belongs to the Convolvulaceae family, commonly known as the morning glory family, and is classified as a vegetable plant. There are several varieties of water spinach, including terrestrial and aquatic types. Water spinach is often cultivated as a complementary food, especially in restaurants and food stalls, where it is frequently used as a key ingredient in many dishes (Pratiwi Baharsyah & Iqbal Suriansyah, 2024).

Packaging is a widely used method for maintaining the freshness and extending the shelf life of food products. The

preservation of agricultural products through packaging plays a crucial role in preventing or minimizing damage by protecting the food contents. Additionally, packaging serves as a barrier against contamination and physical disturbances (Ropikoh et al., 2024). According to Maharani et al. (2021), kale plants are highly perishable and can lose freshness due to transpiration and respiration. Transpiration is a physiological process in which plants release water (H₂O) through evaporation, leading to weight loss, reduced water content, and diminished freshness.

Storing agricultural products at cold or low temperatures can also inhibit respiration, thereby slowing softening, color changes, quality changes, and other chemical processes. However, storing agricultural products in the refrigerator or at cold temperatures can also cause

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cell freezing, because most cells are composed of fluids that can freeze at low temperatures. According to Ambuko et al. (2017), water spinach plants are easily damaged and lose freshness due to transpiration and respiration. Transpiration is a physiological process in which plants release H₂O through evaporation, leading to weight loss, reduced H₂O content, and decreased freshness.

2. Material and Methods

2.1. Research Location

This research was conducted at the Postharvest Complex, Horticultural Research Centre, Malaysian Agricultural Research and Development Institute (MARDI), Serdang, Selangor, at coordinates 2.9911°N, 101.7016 °E, 50 masl. The experiment was designed to evaluate the effect of passive modified atmosphere (MAP) packaging using sealed LDPE film without holes, compared with perforated LDPE film (control), on the postharvest quality of water spinach (*Ipomoea aquatica*). The experiment followed a completely randomized design (CRD) with two packaging treatments and three replications per sampling day.

2.2. Research Plant Materials

Freshly harvested kale shoots at commercial maturity were obtained from local vegetable gardens in Selangor. Only uniform, defect-free samples were selected. Samples were prepared with the roots still attached without any additional treatment. Approximately 3 kg of kale was placed in each packaging unit.

2.3. Research methods

2.3.1. Packaging Treatment and Storage Conditions

Two types of low-density polyethylene (LDPE) packaging films are used:

- T₁ : LDPE without holes (thickness 0.04 mm) - MAP
- T₂ : 1% perforated LDPE (thickness 0.04 mm) – Control

Each package was heat-sealed and stored in a refrigerator maintained at 10 ± 1 °C and 95% relative humidity. Sampling was performed on days 0, 5, and 10 of storage for all analyses.

2.3.2. Determination of Chlorophyll Content

Ascorbic acid (vitamin C) was determined by titration using a 2,6-dichlorophenol-indophenol (DCPIP) solution. Approximately 5 g of the sample was homogenized in 3% metaphosphoric acid and filtered through Whatman No. 1 paper. A 10 mL aliquot of the filtrate was titrated with a standard DCPIP solution until a light pink color persisted for 5 seconds. The results were expressed as mg of ascorbic acid per 100 g of fresh weight (mg 100 g⁻¹ FW).

2.3.3. Color Measurement

The surface color of water spinach leaves was measured using a colorimeter calibrated with a white

standard. CIELAB parameters a* (green–red) and b* (blue–yellow) were recorded at three random points per sample, and the average value was used for statistical analysis.

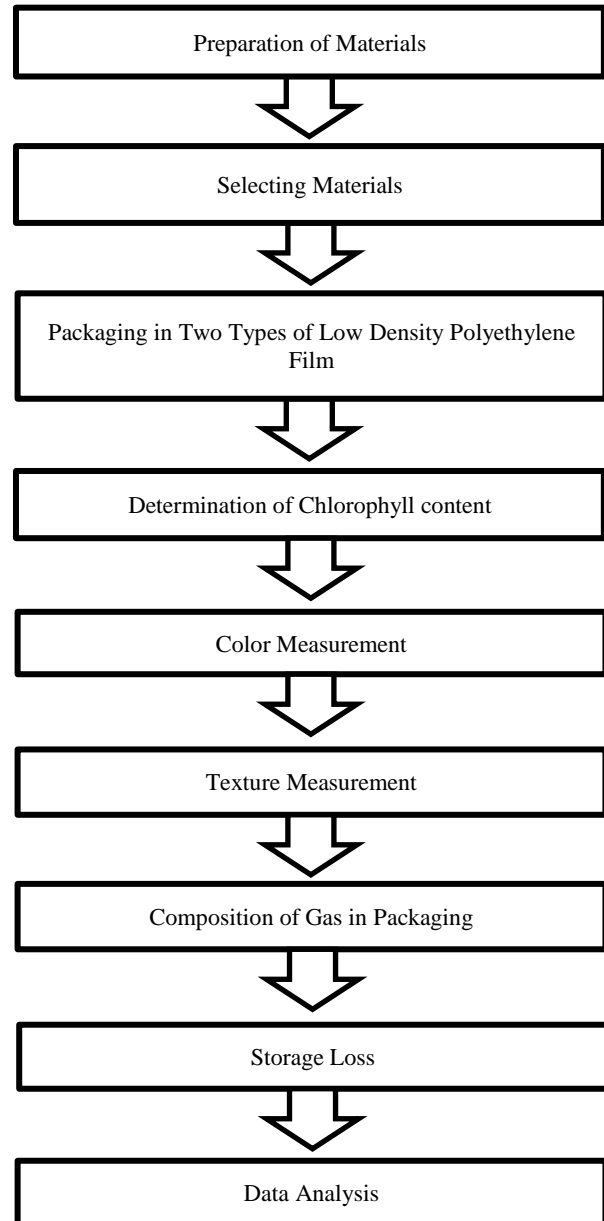


Figure 1. Research flow diagram

2.3.4. Elasticity

The springiness of the leaf stalk was measured using a texture analyzer (Model TA-TX plus Stable Micro Systems, UK) equipped with a knife probe. The testing speed was set at 10 mm s⁻¹, and the maximum force (N) required to penetrate the sample was recorded as springiness (N).

2.3.5. Composition of Gas in Packaging

The concentrations of oxygen (O₂) and carbon dioxide (CO₂) within each package were determined using

a gas chromatograph (PerkinElmer). Gas samples (~10 mL) were taken from each sealed bag using a gas-tight syringe. Measurements were recorded as percentages of O₂ and CO₂ on each sampling day.

2.3.6. Storage Loss

Storage loss (%) was calculated as the proportion of kale rejected due to visual damage (wilt, yellowing, or rot) relative to the initial sample weight on each day of storage.

$$\text{Formula: Storage loss (\%)} = \frac{W_r}{W_0} \times 100$$

where W₀ = initial sample weight, W_r = weight rejected due to damage.

2.4. Data Analysis

Data were analyzed using analysis of variance (ANOVA) in SAS software (Version 94, SAS Institute Inc.,

Cary, NC, USA). Comparisons of means were performed using Duncan's Multiple Range Test (DMRT) at p < 0.05 to determine significant differences between treatments and storage duration.

3. Results and Discussion

Two-way ANOVA (Table 1) showed that among the measured parameters, chlorophyll content, gas composition (O₂ and CO₂), and storage losses were significantly affected by packaging treatment and showed a significant interaction between treatment and time (Table 1). This result indicates that kale responded differently to unperforated and perforated LDPE films during storage. Meanwhile, all quality parameters were significantly affected by storage duration (p < 0.001), indicating that changes progressed with increasing storage time.

Table 1. Summary of two-way ANOVA (p-value) for postharvest parameters of water spinach stored with different packaging treatments.

Source	content chlorophyll	Ascorbic acid content	a* (green-red)	b* (blue-yellow)	elasticity	in O ₂ packaging	CO ₂ packaging	Storage loss %
Day	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)	<0.0001 (***)
Maintenance	0.0014 (**)	0.6903 (NS)	0.8296 (NS)	0.4408 (NS)	0.3914 (NS)	<0.0001 (***)	<0.0001 (***)	0.0004 (***)
Day × Treatment	0.0001 (***)	0.9309 (NS)	0.7518 (NS)	0.3129 (NS)	0.2469 (NS)	<0.0001 (***)	<0.0001 (***)	0.0145 (*)

Note: NS = not significant; * = significant at p < 0.05; ** = significant at p < 0.01; *** = significant at p < 0.001. The interaction effect (Day × Treatment) indicates whether the treatment difference varied across storage days.

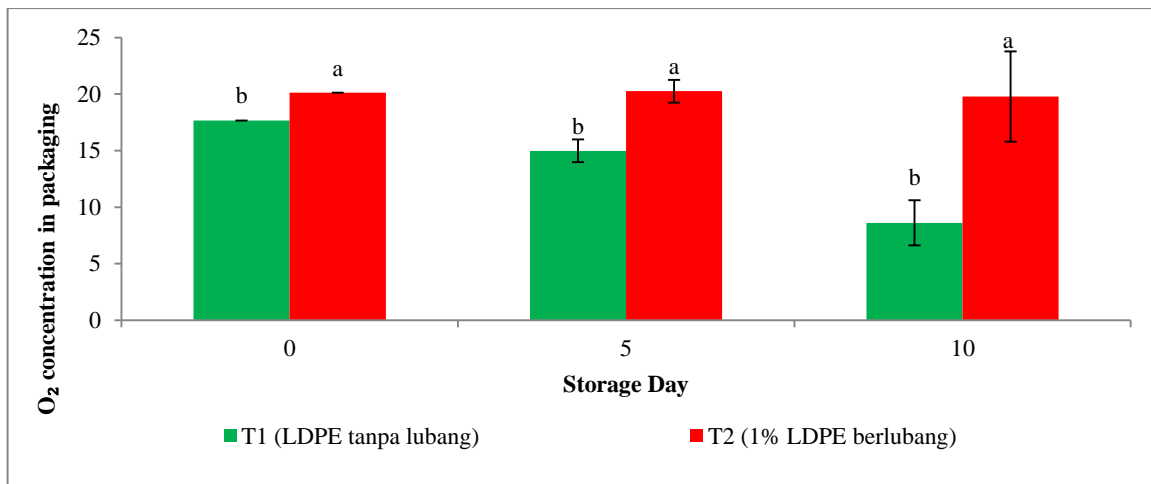


Figure 2. Changes in O₂ concentration in the packaging during storage of kale in LDPE packaging without holes (T₁) and LDPE packaging with 1% holes (T₂) at a temperature of 10 ± 1 °C for up to 10 days.

The image above shows a bar graph depicting the concentration of O₂ in the packaging over storage days. The x-axis shows storage days (0, 5, and 10), while the y-axis shows the O₂ concentration in the package (as a percentage). T1 (Non-perforated LDPE): Marked with a green bar, shows a relatively stable O₂ concentration of

around 15-20% on days 0 and 5, then decreases to around 10% on day 10. T2 (1% perforated LDPE): Marked with a red bar, shows a higher O₂ concentration, around 20% on days 0 and 5, with a marked increase to around 25% on day 10. These data indicate that T2 packaging with 1% perforation increases O₂ concentration over time, whereas

T1 packaging without perforation decreases O₂ concentration. A decrease in O₂ concentration in the packaging can reduce respiration rate and slow aging in kale. However, if the O₂ level is too low (<1%), anaerobic conditions can develop, triggering fermentation and

causing unpleasant odors and quality degradation. (Qian et al., 2022) Therefore, the O₂ concentration needs to be regulated within the optimal range of 3–6%, with CO₂ levels between 3–10% to maintain leaf freshness without triggering physiological damage.

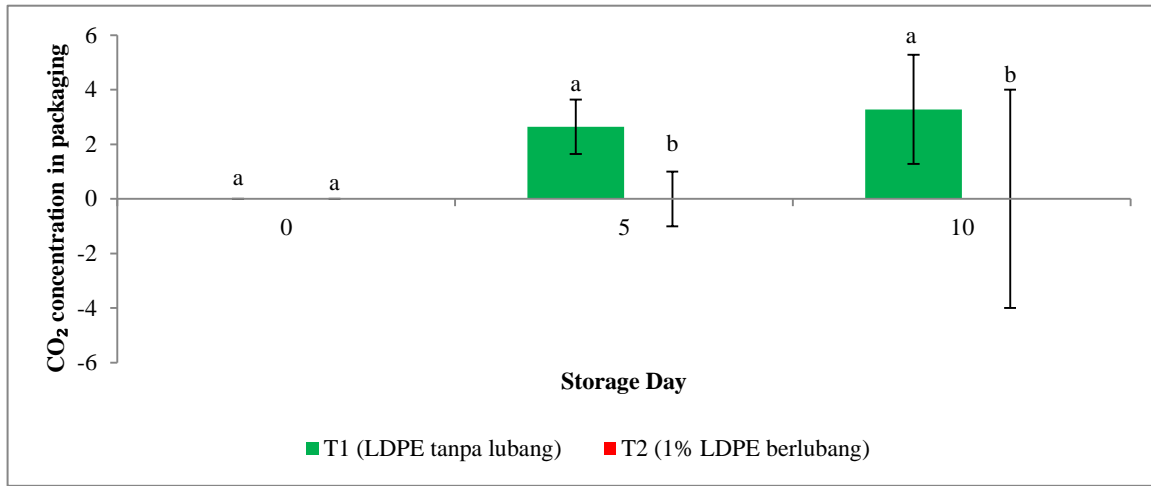


Figure 3. Changes in CO₂ concentration in the packaging during storage of kale in LDPE packaging without holes (T₁) and LDPE packaging with 1% holes (T₂) at a temperature of 10 ± 1 °C for up to 10 days.

In the bar graph depicting the concentration of CO₂ in the packaging against the days of storage. The x-axis shows the storage days (0, 5, and 10), while the y-axis shows the CO₂ concentration in the package (as a percentage). T1 (Non-perforated LDPE): Marked with a green bar, shows a stable CO₂ concentration near 0% on days 0 and 5, with a slight increase to around 2-4% on day 10. T2 (1% perforated LDPE): Marked with a red bar, but not visible in the data above and therefore not significant. The data show that the T1 packaging without perforation tends to maintain a low CO₂ concentration initially, with a small increase over time, which may be due to the

material's respiratory activity within the package, and perforation at T2. According to Karbon et al. (2025), the average decrease in CO₂ was 4% after 2 hours, 8% after 4 hours, 15% after 6 hours, 22% after 12 hours, and 32% after 24 hours compared to the initial conditions (pre-test). The longer the exposure, the greater the decrease in CO₂, with the most significant effect at 24 hours (a decrease of up to 305 ppm from 1,345 ppm). while at 28 °C, the CO₂ concentration rapidly increased and then decreased to a constant value.

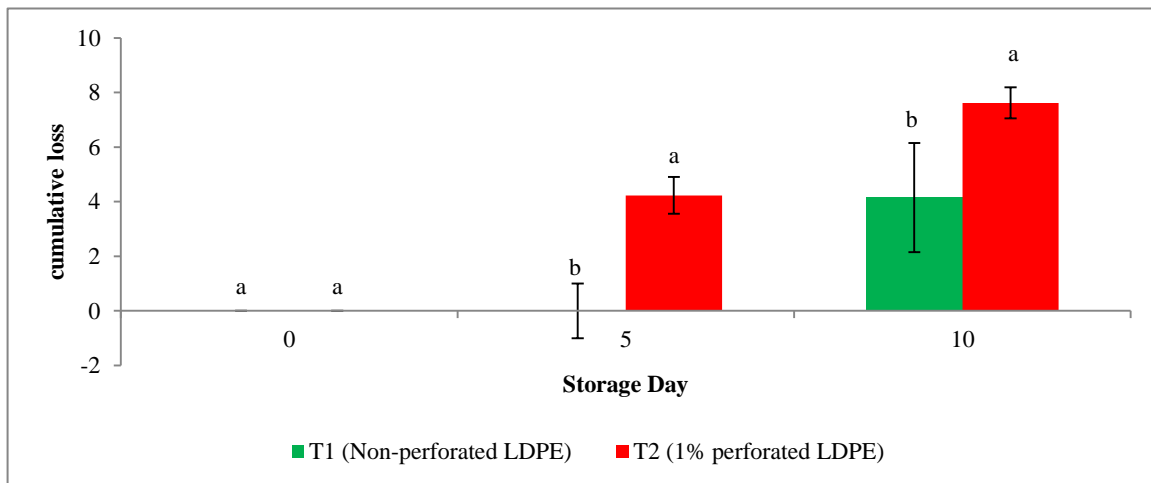


Figure 4. Storage losses of kale stored in non-perforated (T₁) and 1% perforated (T₂) LDPE packaging at 10 ± 1 °C for up to 10 days. Bars represent mean ± SE (n = 3). Different letters within the same day indicate significant differences (p < 0.05) based on the DMRT test.

After being removed from storage, samples were unpacked and immediately evaluated for physical condition.

Visual observation revealed no significant differences in leaf color between treatments at any evaluation point. However, the decline in kale quality generally began with wilting, followed by yellowing, and finally decay. Damaged samples from each treatment were separated and weighed to calculate the cumulative percentage of rejected samples, expressed as storage losses. Cumulative storage losses increased significantly with storage duration and

were consistently higher at T_2 than at T_1 . A significant Day \times Treatment interaction indicated that the treatment effect became more pronounced at later storage stages. Water spinach stored in non-perforated LDPE packaging (T_1) recorded lower losses after Day 10, consistent with reduced wilting symptoms. According to Hoffmann et al. (2021), water loss in vegetables and fruits can occur through evaporative heat from respiration.

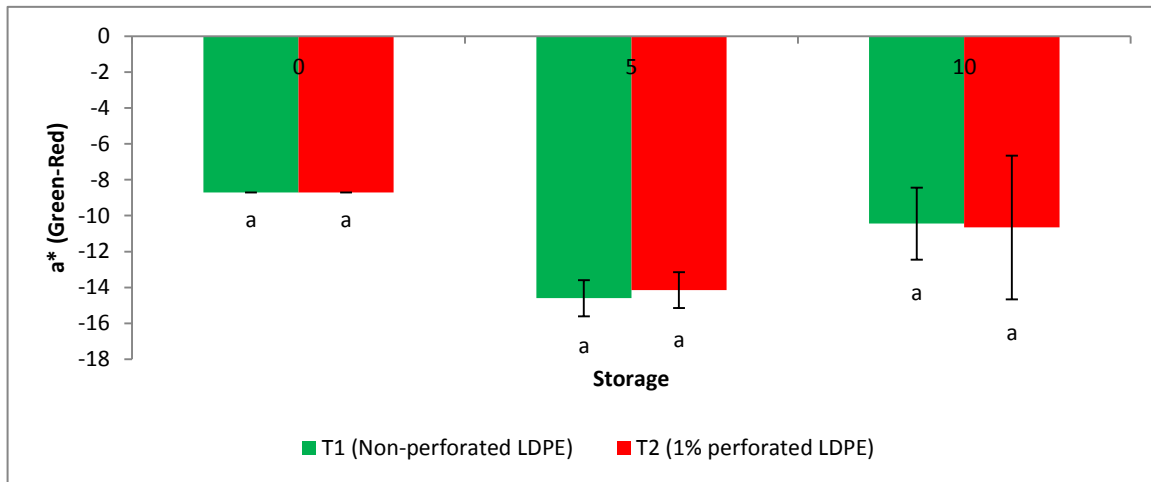


Figure 5. Changes in the a^* (green–red) value of water spinach leaves stored in LDPE packaging without holes (T_1) and 1% holes (T_2) at a temperature of 10 ± 1 °C for up to 10 days.

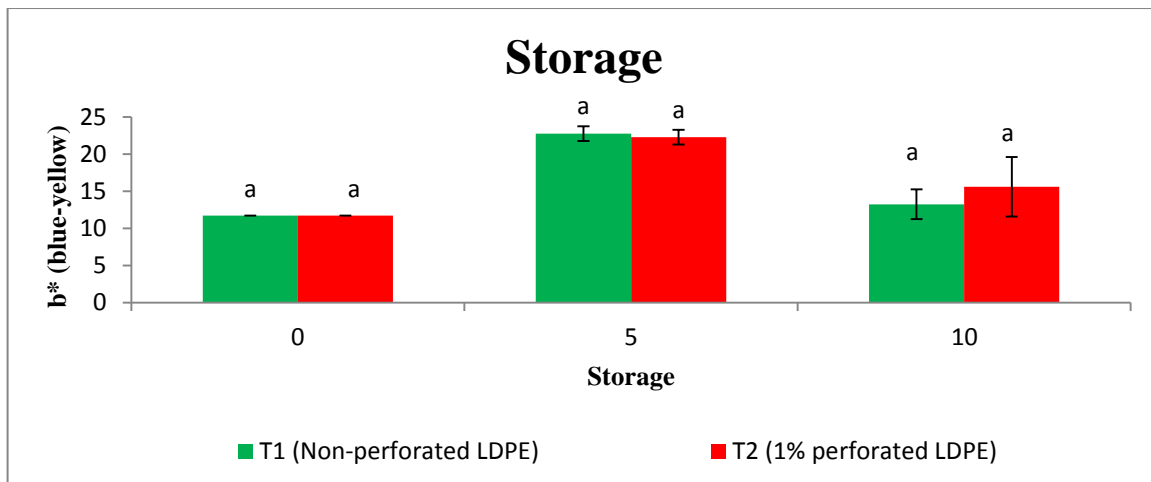


Figure 6. Changes in b^* value (blue–yellow) in water spinach stored in LDPE packaging without holes (T_1) and LDPE packaging with 1% holes (T_2) at a temperature of 10 ± 1 °C for up to 10 days.

Based on CIELAB color assessment, both a^* and b^* color indices were significantly affected by storage duration, but showed no significant effect of treatment or their interaction. The a^* value became less negative, while the b^* value increased over time, indicating a gradual shift in leaf color from green to yellow. This change reflects the progressive loss of chlorophyll and the appearance or synthesis of carotenoid pigments during senescence. The absence of a treatment effect indicates that the difference in gas composition between the two types of LDPE packaging was insufficient to affect surface color development during

the first 10 days of storage. In general, the a^* coordinate represents the degree of greenishness (negative values) or reddishness (positive values), while the b^* coordinate represents the degree of bluishness (negative values) or yellowishness (positive values) on the color wheel. Based on research results, color changes during storage of various horticultural products for 14 days, the fastest color change occurred in kale with brown skin and blackish spots. In general, the longer a horticultural product is stored, the darker its color. Color changes can be caused by humidity, storage environment temperature, or the process of

chlorophyll decomposition in green vegetables, which can cause a color change from green to yellow or brownish.

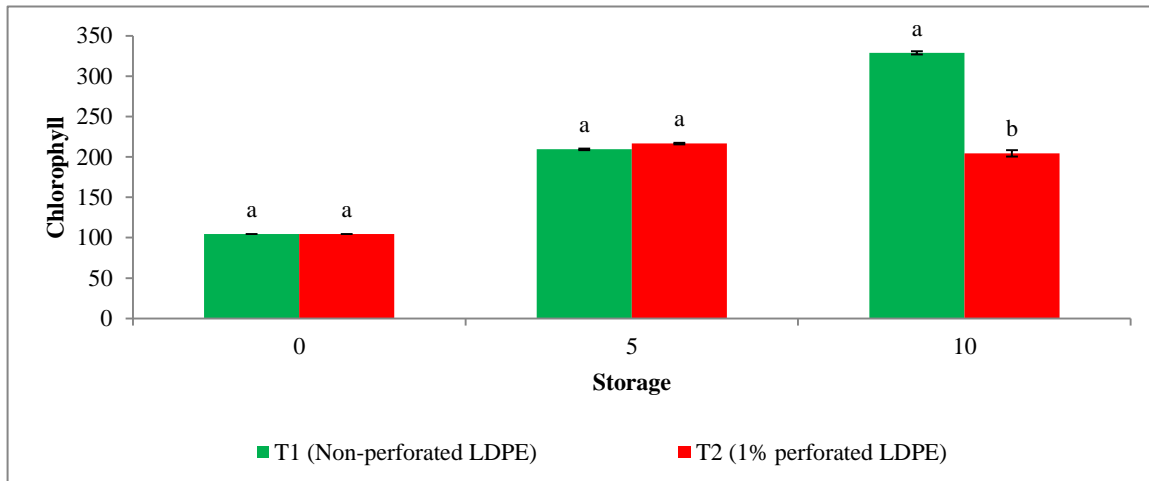


Figure 7. Changes in chlorophyll content of water spinach stored in LDPE packaging without holes (T_1) and 1% holes (T_2) at 10 ± 1 °C for up to 10 days. Bars represent mean \pm SE ($n = 3$). Different letters within the same day indicate significant differences ($p < 0.05$) according to DMRT.

An unusual trend was observed, with chlorophyll content increasing progressively from Day 0 to Day 10, likely reflecting the accumulation or concentration of chlorophyll degradation products during storage. Although measured chlorophyll values increased until Day 10, this pattern is likely a concentration effect due to water loss and spectral interference from degradation products such as pheophytin and chlorophyllide, which overlap in absorption with chlorophyll a and b (Chiarenza et al., 2024). A clear treatment effect was observed at the final stage of storage (Day 10), where kale packed in unperforated LDPE (T_1)

retained a higher chlorophyll content than that packed in perforated LDPE (T_2). This result suggests that the tightly sealed LDPE creates an environment with low oxygen and high carbon dioxide levels, which slows chlorophyll degradation and maintains greener leaves. In contrast, perforated LDPE allows continuous gas exchange, which accelerates pigment loss. In addition, increasing CO_2 levels can further enhance chlorophyll retention, as high CO_2 levels have been shown to inhibit chlorophyll degradation (Wei et al., 2021).

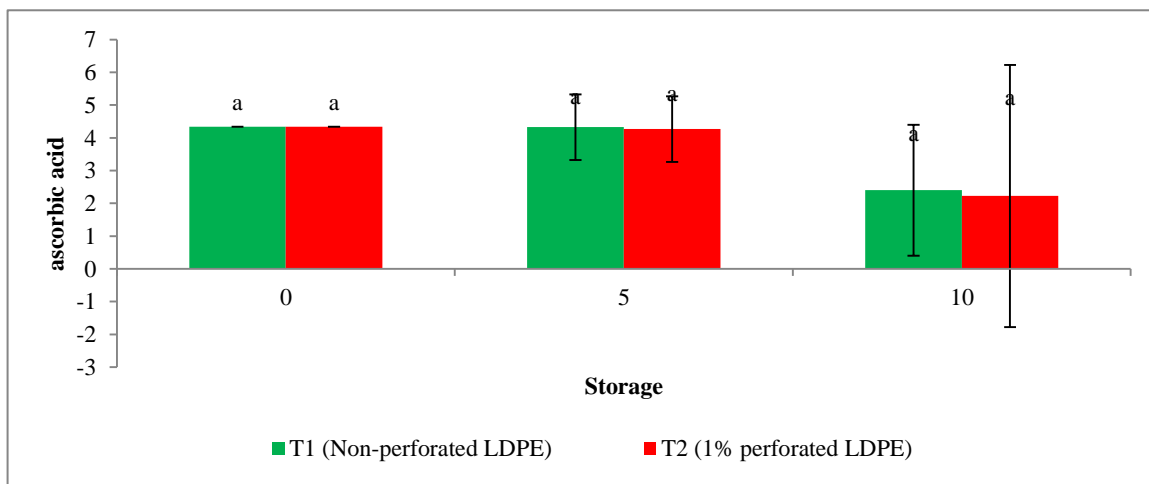


Figure 8. Changes in ascorbic acid content of water spinach during storage in LDPE packaging without holes (T_1) and 1% holes (T_2) at a temperature of 10 ± 1 °C for up to 10 days. Bars represent the mean \pm SE ($n = 3$).

In general, ascorbic acid is a major metabolite in plants that functions as an antioxidant, protecting plants from oxidative damage caused by aerobic metabolism, photosynthesis, and various pollutants. Ascorbate is also a cofactor for several hydroxylase enzymes (e.g., prolyl hydroxylase) and violaxanthin de-epoxidase. In this study,

ascorbic acid content decreased significantly ($p < 0.001$) during storage, regardless of packaging treatment. This decrease was caused by oxidative degradation during storage, a common phenomenon in green leafy vegetables. However, there was no significant difference between treatments, indicating that both films provided comparable

protection under the same cold storage conditions. According to Jessica Emai & Radix Astadi Praptono Jati (2024), the vitamin C (ascorbic acid) content in kale decreased slightly over 7 days of storage, but the thickness of the PVC packaging (0.010 cm or 0.013 cm) did not significantly affect the change. The initial vitamin C value

was around 5–6 mg/100 g, and after 7 days it decreased to 4–5 mg/100 g (an average decrease of 15–20%). This decrease was caused by natural respiration and oxidation processes, although the plastic packaging helped slow the loss compared to no packaging.

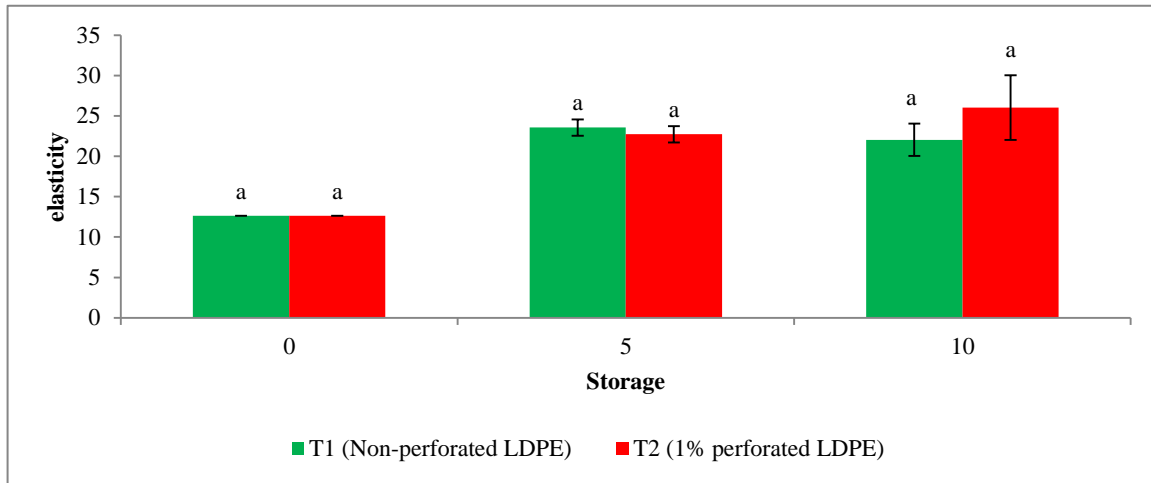


Figure 9. Firmness of water spinach stored in LDPE packaging without holes (T_1) and 1% holes (T_2) at a temperature of 10 ± 1 °C for up to 10 days.

The figure above shows the level of satiety (in percentage) as a function of storage days. The x-axis shows storage days (0, 5, and 10), while the y-axis shows satiety value. T1 (Non-perforated LDPE) Marked with a green bar, shows a satiety of about 15% on day 0, increasing to about 22-25% on day 5, and slightly decreasing to about 20% on day 10. T2 (1% perforated LDPE): Marked with red bars, indicating similar firmness to T1 at around 15% on day 0, increasing to around 22-25% on day 5, and jumping to around 25-30% on day 10. These data indicate that both packaging types increased in firmness until day 5, with T2 showing a greater increase on day 10. The perforations in T2 likely allowed for greater moisture absorption, thus

increasing firmness compared to T1. Leaf firmness increased gradually over storage ($p < 0.001$), indicating a significant effect of storage duration but no difference between treatments. The observed increase in leaf firmness likely reflects physiological water loss and decreased cell elasticity, rather than an improvement in textural quality. As storage progressed, the tissue became harder and less tender, indicating a progressive decline in leaf freshness regardless of packaging treatment. According to Kumara and Beneragama (2020), the firmness of sponges continued to increase during storage due to respiration and transpiration, which result in water loss. Water loss is the main cause of wilting.



Figure 10. Results of Treatment 1.



Figure 11. Results of Treatment 2.

4. Conclusion

Based on the results of this study comparing the T1 and T2 treatments, the T1 treatment is superior. Observations up to the tenth day indicate that the percentage loss in T1 is slightly lower than in T2. This result is due to T1's completely sealed packaging, which limits kale's exposure to air and helps it stay fresh longer.

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