



## Research article

Effects of active and passive modified atmosphere packaging on biochemical properties of cut *Dendrobium* orchid flowers

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## ABSTRACT

Optimal storage conditions are essential to preserving the quality of postharvest produce and cut flowers during transportation, especially to overseas destinations. As a result, this research investigated the effects of active and passive modified atmosphere packaging (MAP) on the biochemical properties of cut *Dendrobium* Sonia orchid flowers. In active MAP, the orchid flowers were wrapped in MAP plastic film filled with 5 % carbon dioxide (CO<sub>2</sub>) and 2 % oxygen (O<sub>2</sub>). Meanwhile, in passive MAP the flowers were stored inside MAP plastic film without filling with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub>. The experimental MAP plastic films were polyethylene, low density polyethylene, high density polyethylene, polypropylene, and polyvinyl chloride films. The biochemical parameters included storage life, respiration rate, ethylene production, internal O<sub>2</sub> and CO<sub>2</sub>, anthocyanin content, protein degradation, and electrolyte leakage. The results showed that the average storage life of orchid flowers under passive MAP condition was 9–15 days, depending on the plastic film types. The longest storage life of 15.66 days was achieved with polypropylene film. The storage life of orchid flowers in active MAP was 9.33 days on average. Without MAP (control), the storage life was 7 days under normal atmosphere condition (0.03 % CO<sub>2</sub> and 21 % O<sub>2</sub>). The experiments also demonstrated that MAP efficiently reduced respiration rate, ethylene production, anthocyanin degradation, protein degradation, and electrolyte leakage. Unlike existing research on MAP which focused primarily on extending the shelf life of fresh produce or cut flowers, this study comparatively investigated the biochemical properties of cut orchid flowers stored in MAP environment, in addition to the storage life.

## 1. Introduction

Orchids (*orchidaceae*) are a family of flowering plants that can grow in various geographical areas, from humid rain forests to mountainous areas. Thailand can grow different species of orchids and is a leading exporter of cut *Dendrobium* orchid flowers (Poonsri, 2017). According to the Department of International Trade Promotion (2019), the export of *Dendrobium* orchid flowers from Thailand accounted for 75 % of the global market share, followed by Singapore (10 %), South Korea (3 %) and others (12 %).

The annual export value of Thailand's *Dendrobium* orchid flowers was USD 88.57 million (Department of International Trade Promotion, 2019). Of the total export value, cut *Dendrobium* Sonia orchid flower varieties, including *Sonia Bom 17*, *Sonia Bom 17 mutation*, *Sonia Bom K*, *Sonia Bom Joe*, accounted for 90 % of the total export value and 10 % belonged to other orchid cultivars (Department of Agriculture, 2019).

Due to their economic significance, this research thus focused on cut *Dendrobium* Sonia orchid flowers.

In exportation of cut flowers, the choice of storage and packaging plays a crucial role in preserving the quality of flowers. Petal discoloration, tissue browning, loss of flower parts, wilting, and senescence shorten the vase life and lower the commercial value of cut flowers. Undesirable postharvest physicochemical changes during storage could be delayed by modifying carbon dioxide (CO<sub>2</sub>) and oxygen (O<sub>2</sub>) concentrations inside the packaging.

According to Poonsri (2017), higher CO<sub>2</sub> and lower O<sub>2</sub> storage atmosphere lowered ethylene production and extended the life of fresh produce. In addition, higher CO<sub>2</sub> and lower O<sub>2</sub> hindered ethylene production inside the storage and delayed flower senescence (Andrew et al., 2009; Burg and Burg, 1965; Dilley, 2006). Modified atmosphere packaging (MAP) has been effectively used to delay the postharvest physicochemical changes of fresh produce and cut flowers (Kassim and Workneh, 2020; Bishop et al., 2007).

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MAP effectively prolonged the vase life of *Gerbera*, *Lilium* and *Rosa* cultivars, compared with those stored without MAP (De Pascale et al., 2005). Patel and Singh (2009) studied the effects of passive MAP and storage temperature (5, 10, and 15 °C) on the flower quality and vase life of *Gerbera* flowers using polypropylene, high density polyethylene, and low density polyethylene films. The results showed that the flowers stored in polypropylene and high density polyethylene passive MAP at 5 °C had significantly lower weight loss and scape bending. The polypropylene and high density polyethylene MAP also delayed petal senescence and enhanced the vase life.

Sealed gas-impervious cellophane MAP delayed the rates of flower opening and extended the vase life of cut rose flowers due to high internal CO<sub>2</sub> and relative humidity (RH) (Hauge et al., 1947). Zeltzer et al. (2001) experimented with modified atmosphere (MA) plastic-lined boxes storing cut rose cultivars at 2 °C, and reported that the fresh weight of the flowers in the MA boxes was three times more than those in the conventional box. The MA boxes also delayed the senescence of flowers and leaves. The results could be attributed to the ethylene inhibitory effect of higher CO<sub>2</sub> and lower O<sub>2</sub> (Defilippi et al., 2006).

Christian et al. (2016) investigated the effect of MAP (higher CO<sub>2</sub> and lower O<sub>2</sub>) on the postharvest life of *Red Globe* table grapes and the rachis appearance stored at 0 °C. The results showed that MAP significantly delayed rachis browning after 90 days storage, compared with those stored without MAP.

Villalobos et al. (2018) studied the effect of passive MAP on the volatile compound profile of postharvest figs; and reported that MAP delayed changes in the compound profile while preserving the fig flavor. Wang et al. (2015) investigated the effects of MAP on the quality of postharvest *Lapins* and *Skeena* sweet cherry cultivars after long-distance ocean freight shipment, including flavor loss, off-flavor development, skin darkening, pedicel browning, pitting, and decay. The results showed that MAP effectively preserved the quality attributes of the fruits.

However, the focus of existing studies on MAP storage is to prolong the storage life of fresh produce and/or cut flowers and preserve the physical appearance and tastes or flavors. To the best of the author's knowledge, there exists no research that investigates the effects of MAP on the biochemical properties in cut flowers.

As a result, this research comparatively investigated the effects of active and passive MAP on the biochemical properties of cut *Dendrobium* Sonia orchid flowers. The experimental MAP films included polyethylene, low density polyethylene, high density polyethylene, polypropylene, and polyvinyl chloride films. In active MAP, the orchid flowers were wrapped in MAP plastic film filled with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub>. In passive MAP, the flowers were retained inside MAP plastic wrap (without filling with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub>). The storage temperature and RH were 13 °C and 95 %. The biochemical parameters under study were the storage life of orchid flowers, respiration rate, ethylene production, internal O<sub>2</sub> and CO<sub>2</sub>, anthocyanin content, protein degradation, and electrolyte leakage. In addition, the results in the active and passive MAP environments were compared with those of orchid flowers stored under normal atmosphere condition (0.03 % CO<sub>2</sub> and 21 % O<sub>2</sub>) without MAP. The novelty of this research lies in the use of active and passive MAP to investigate the biochemical properties in cut *Dendrobium* orchid flowers, unlike existing research which focused primarily on fresh vegetables and fruits.

## 2. Materials and methods

In this research, *Dendrobium* Sonia orchid flowers were obtained from a plantation in *Pathumthani* province of Thailand. Prior to experiments, the flowers were thoroughly washed in tap water, and the flower stem ends were trimmed and inserted into plastic tubes filled with distilled water (30 mL). Figure 1 depicts cut *Dendrobium* Sonia orchid flowers before wrapping in MAP.

This research experimented with five MAP film types: polyethylene (PE), high density polyethylene (HDPE), low density polyethylene



Figure 1. Cut *Dendrobium* Sonia orchid flowers prior to wrapping with MAP.

(LDPE), polyvinyl chloride (PVC), and polypropylene (PP) films. The dimensions of the MAP films were 22 × 55 cm (W × L) and 24 μm in thickness. The experiments with different film types (both active and passive MAP) and under normal atmosphere condition (control) were carried out in triplicate with 40 inflorescences per replicate. The orchid flowers in the active MAP plastic films were filled with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub> using a gas mixer (MAP Mix Provectus-Dansensor MAP Check 3, Denmark) and heat sealed prior to placing in the cardboard boxes. Meanwhile, those in passive MAP plastic films were heat sealed (without filling additional CO<sub>2</sub> and O<sub>2</sub>) and then placed in the cardboard boxes. The wrapping of the flowers with plastic films was carried out at room temperature (25 °C). The flowers under normal atmosphere condition were placed in the cardboard boxes (without MAP). The storage temperature and RH were 13 °C and 95 %, and the flowers were retained in the dark, following the protocol of Thailand's Department of International Trade Promotion. According to Poonsri (2015), the optimal storage conditions of *Dendrobium* orchid flowers were 13 °C and 95 % RH. The storage life and biochemical properties of the orchid flowers were assessed every three days for the entire experimental period of 15 days and results compared.

### 2.1. Storage life

The vase life of freshly cut *Dendrobium* Sonia orchid flowers at room temperature (25 °C and 60 % RH) was first determined and used as the reference. In this study, the vase life of freshly cut orchid flowers was seven days, given that over half of the flowers wilted. In the storage life assessment, the orchid inflorescences stored under normal atmosphere condition (control) and those wrapped in active and passive MAP were unpacked and placed in vases filled with distilled water at room temperature (25 °C and 60 % RH). The storage life assessment was carried

out on days 3, 6, 9, 12, and 15. The orchid flowers were visually inspected daily by a panel of 10 specialists with knowledge of flowers and the observation results compared with the freshly cut orchid flowers (reference). The storage life of the experimental orchid flowers (control and active and passive MAP) ended if the flowers lasted less than seven days at room temperature due to shorter vase life of the freshly cut orchid flowers (<7 days).

## 2.2. Respiration rate

The respiration rate analysis was carried out by gas chromatography, following Watkins et al. (2000) with minor modifications, whereby 1 g of orchid petals was transferred to a hermetic flask and retained for 1 h at room temperature. Gas samples (1 mL) were collected through silicone septum using a syringe and analyzed by a gas chromatography machine (Agilent Technologies; 6820, USA) equipped with a capillary column (2 m) at 100 °C. Hydrogen (100 kPa) was used as the carrier gas with a thermal conductivity detector. The respiration rate of orchid flowers was expressed as mg CO<sub>2</sub> kg<sup>-1</sup> h<sup>-1</sup>.

## 2.3. Ethylene production

The ethylene production was measured every three days for 15 days using an ethylene analyzer (ICA; 56, UK), and results were reported in parts per million (ppm).

## 2.4. Internal O<sub>2</sub> and CO<sub>2</sub>

In the analysis of internal O<sub>2</sub> and CO<sub>2</sub> concentrations, 1 mL of gas samples were collected through silicone septum using a syringe and analyzed for 35 s using a gas analyzer (PBI-Dansensor Checkmate II, Denmark). The internal O<sub>2</sub> and CO<sub>2</sub> concentrations were expressed as a percentage.

## 2.5. Anthocyanin content

Anthocyanin is water-soluble vacuolar pigments that play an essential role in the color and physical attractiveness of *Dendrobium* orchid flowers (Asen et al., 1973; Kader, 2002). According to Zhang et al. (2001), increased vacuolar pH caused anthocyanins to degrade and induced senescence.

In the anthocyanin extraction, 0.5 g crushed orchid petal was submerged in 25 mL acidic ethanol (1.5 M HCl in 95 % ethanol, 15:85 v/v) at 4 °C overnight. After 24 h, the solution was filtered with Whatman No.1 paper, and the volume was adjusted to 100 mL by adding acidic ethanol (Rangana, 1986). The analysis of anthocyanin content was carried out using a spectrophotometer at 535 nm wavelength (Labomed; SPECTRO 23, USA), and the measurements were converted into mg of anthocyanin per 100 g fresh weight (Eqs. (1) and (2)).

$$\text{Total Absorbance} = \frac{OD \times V \times 100}{W} \quad (1)$$

$$\text{Total anthocyanin content} = \frac{\text{Total Absorbance}}{98.2} \quad (2)$$

where OD is the absorbance value at 535 nm, V is the solvent volume, W is the weight of crushed orchid petal, and 98.2 is a constant.

## 2.6. Protein degradation

Prior to the protein degradation analysis, orchid petals immersed in liquid N<sub>2</sub> were first ground and mixed in 5 mL extraction medium which contained 50 mM Tris HCl pH 7.6, 2 mM disodium

ethylenediaminetetraacetate, 2 mM dithiothreitol, and 10 mM MgCl<sub>2</sub>. The ground orchid petals in the mixture was then incubated for 15 min at room temperature (25 °C) before centrifugation at 17,000 g for 20 min at 4 °C. The supernatant was collected and 2 mL extraction medium was added to the pellet prior to a second extraction.

In the second extraction, the mixture (the pellet in 2 mL extraction medium) was incubated for another 15 min at room temperature and subsequently centrifuged at 17,000 g for 20 min at 4 °C. The supernatant (soluble protein) of the second extraction was combined with the supernatant from the first extraction. Furthermore, 7 mL of 0.1 N NaOH was added to the pellet from the second extraction and incubated at 80 °C for 24 h in order to extract insoluble protein. In this study, total protein included soluble and insoluble proteins.

The total protein (both soluble and insoluble proteins) was analyzed for protein degradation by using the spectrophotometer (Labomed; SPECTRO 23, USA) at 595 nm wavelength with bovine serum albumin used as the standard protein assay (Bradford, 1976). In the analysis, 50 μL of protein was pipetted into 1.5 mL microcentrifuge tube which contained 200 μL protein buffer, and 1 mL of 0.0125 % Coomassie Brilliant Blue (CBB) was added. The mixtures were retained at room temperature (25 °C) for 15 min before measurement of the protein content, which was expressed as mg g<sup>-1</sup> fresh weight.

## 2.7. Electrolyte leakage

In the analysis of electrolyte leakage, orchid petals were cut into 1-cm-diameter discs (approximately 1 g in weight) before thoroughly washing with deionized distilled water and left to dry on tissue paper. The orchid petal discs (10 discs per experiment) were transferred to centrifuge tubes filled with 30 mL 0.4 M mannitol solution and shaken using refrigerated centrifuge at 100 cycles per minute (Hettich Universal 320R, Germany) (McCollum and McDonald, 1991).

The mixture was subsequently incubated for 3 h at room temperature (25 °C), and the initial reading of electrolyte leakage (i.e., electrical conductivity) was determined by using a conductivity meter (HANNA instruments; EC 214, Portugal). The mixture was then placed in an autoclave (Gemmy Industrial; Speedy Autoclave Vertical Type HL341, Taiwan) and treated for 30 min at 121 °C and 15 psi before allowing to cool to room temperature. The final reading of electrolyte leakage (i.e., total electrical conductivity) was measured. The percentage of electrolyte leakage was calculated as the ratio of initial reading to final reading (Eq. (3)).

$$\text{Electrolyte leakage (\%)} = \frac{\text{Initial conductivity reading}}{\text{Total conductivity reading}} \times 100 \quad (3)$$

## 2.8. Statistical analysis

Tukey's multiple comparison test was used to determine statistical differences in the biochemical properties between the orchid flowers wrapped in active and passive MAP and the control using Statcel3 software program (OMS, Tokyo, Japan), given the 5 % significance level.

## 3. Results and discussion

This section compares the experimental results of cut *Dendrobium* Sonia orchid flowers stored in active and passive MAP with those under normal atmosphere condition without MAP (control). Because of the longest storage life of orchid flowers wrapped in PP-film passive MAP, the descriptive discussion thus emphasizes the experimental results of passive and active MAP with PP film type, in comparison with those under normal atmosphere condition (control). Meanwhile, the comparative results between the active and passive MAP of other film types and the control were presented graphically and in table format.



### 3.1. Storage life

The average storage life of cut orchid flowers wrapped inside PE, HDPE, LDPE, PP, and PVC films (passive MAP) were 12.33, 12.33, 12.33, 15.66, and 13.00 days, respectively. The average storage life of orchid flowers in active MAP filled with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub> was 9.33 days. The storage life of orchid flowers under normal atmosphere condition (control) was only 7.67 days. Table 1 tabulates the storage life of cut *Dendrobium* Sonia orchid flowers with and without MAP.

The results showed that passive and active MAP significantly increased the storage life of orchid flowers ( $p < 0.05$ ), compared with those under normal atmosphere condition (control). With passive MAP, the orchid flowers wrapped in PP packaging had the longest average storage life (15.66 days). This could be attributed to increased CO<sub>2</sub> and decreased O<sub>2</sub> inside PP packaging (Figures 5 and 6). Higher CO<sub>2</sub> and lower O<sub>2</sub> reduced the respiration rate and ethylene production (Yahia and Singh, 2009). According to Andrew et al. (2009); Poonsri (2020), passive MAP, together with low temperatures, could effectively extend the shelf life of cut flowers (Figure 2).

With active MAP, the average storage life of the orchid flowers was 9.33 days, which was significantly different from those stored in passive MAP ( $p < 0.05$ ). The shorter storage life could be attributed to elevated CO<sub>2</sub> concentrations inside active MAP after 9 days from 5 % to between to 8.65–10.32 %, which were significantly higher than those inside passive MAP (Figure 6). According to Kader (2002), elevated CO<sub>2</sub> induced the physiological breakdown of fresh produce. Mitcham and Shelton (1997) documented that *Dendrobium* orchid flowers exposed to high CO<sub>2</sub> concentrations had shorter storage life than the air-stored flowers. Yahia and Singh (2009) reported that tropical crops were sensitive to high CO<sub>2</sub> concentrations (around 10 %).

### 3.2. Respiration rate

The respiration rates of the control (orchid flowers stored under normal atmosphere condition) were significantly higher than those retained in active and passive MAP ( $p < 0.05$ ), as shown in Figure 3. The orchid flowers in PP-film passive MAP had the lowest respiration rates due to higher CO<sub>2</sub> and lower O<sub>2</sub> concentrations (Figures 5 and 6). The higher CO<sub>2</sub> concentrations in active and passive MAP reduced the respiration rates of the orchid flowers, when compared with those stored under normal atmosphere condition.

Under the normal atmosphere (control), the respiration rate steadily decreased and reached 21.00 mg CO<sub>2</sub>/kg/h after 9 days (when the vase life was shorter than seven days). With PP-film passive MAP, the respiration rate of orchid flowers wrapped in PP film steadily decreased to 12.22 mg CO<sub>2</sub>/kg/h on day 12 and slightly increased to 13.32 mg CO<sub>2</sub>/

kg/h at termination (i.e., day 15). The respiration rate then decreased. The results indicated that the passive MAP condition effectively decreased the respiration rates of orchid flowers.

The lower respiration rates could be attributed to the modified atmosphere inside the packaging (higher CO<sub>2</sub> and lower O<sub>2</sub>) due to the semi-permeability of plastic films (Poonsri, 2020). According to Andrew et al. (2009), higher CO<sub>2</sub> and lower O<sub>2</sub> concentrations, together with low temperatures, reduced the respiration rates in carnations and roses. In addition, MAP reduced the respiration rates of cut *Dendrobium* cv. Red Bomjo (Brandes and Zude-Sasse, 2019; Poonsri, 2017) and Big White Jumbo (Uthairatanakij et al., 2010), cut gerbera flowers (Patel and Singh, 2009), cut carnation flowers and cut Gladiolus flowers (Andrew et al., 2009).

### 3.3. Ethylene production

The ethylene production rates of orchid flowers retained in active and passive MAP were significantly lower than under the normal atmosphere condition ( $p < 0.05$ ), as shown in Figure 4. The ethylene production of the control steadily increased and reached 13.22 ppm on day 6, which is the level of ethylene that induces wilting, abscission and senescence (Rattanapanon and Boonyakiat, 2013; Poonsri, 2020). Due to the excessive ethylene levels, the average storage life of orchid flowers under the normal atmosphere was only 7.67 days. Meanwhile, the ethylene production of the orchid flowers in PP-film passive MAP gradually increased and reached 7.95 ppm on day 12. Due to the gradual ethylene synthesis, the orchid flowers wrapped in PP-film passive MAP lasted 15.66 days on average. The results showed that MAP effectively retarded ethylene production, giving rise to longer storage life for the orchid flowers.

The results could be attributed to higher CO<sub>2</sub> inside MAP, which attached itself to the receptor sites of plant tissues. Specifically, CO<sub>2</sub> competed for the receptor sites with ethylene (Poonsri, 2020). In addition, the semi-permeability of plastic films altered the internal atmosphere (higher CO<sub>2</sub> and lower O<sub>2</sub>), which reduced the ethylene production and ethylene action (Brandes and Zude-Sasse, 2019; Kader, 2002; Andrew et al., 2009; Poonsri, 2021). Specifically, the ethylene production was reduced as low O<sub>2</sub> inhibited the conversion of 1-aminocyclopropane-1-carboxylic acid (ACC) into ethylene (Poonsri, 2020).

### 3.4. Internal O<sub>2</sub> and CO<sub>2</sub>

In Figures 5 and 6, the O<sub>2</sub> and CO<sub>2</sub> concentrations under normal atmosphere condition (control) remained relatively unchanged from day 0 (20.80 % and 0.11 % for O<sub>2</sub> and CO<sub>2</sub>) to day 9 (21.10 % and 0.10 %). The internal O<sub>2</sub> and CO<sub>2</sub> inside passive MAP were 20.00–21.00 % and 0.10–0.11 % (at day 0), and after 12 days, they were 19.43–20.20 % and 0.46–0.90 %. Meanwhile, the initial internal O<sub>2</sub> and CO<sub>2</sub> in active MAP were 2.08–2.41 % and 5.08–5.32 % (day 0) and increased to 5.77–10.31 % and 8.65–10.32 % after nine days. The findings corresponded to the respiration rates.

Specifically, with PP-film passive MAP, the internal O<sub>2</sub> gradually decreased from 20.60 % (day 0) to 19.60 % (day 15), while the internal CO<sub>2</sub> concentration increased from 0.10 % (day 0) to 0.90 % (day 15). With PP-film active MAP, the O<sub>2</sub> and CO<sub>2</sub> concentrations increased from 2.31 % and 5.08 % (day 0) to 6.45 % and 10.32 % (day 9). The findings were consistent with the lower respiration rates and lower ethylene production.

The results also indicated that sealed packaging or MAP lowered the gas exchange between internal and external atmospheres, leading to higher CO<sub>2</sub> and lower O<sub>2</sub> inside the packaging (Kader, 2002; Robertson, 2006; Andrew et al., 2009; Upasen and Wattanachai, 2018). The O<sub>2</sub> and CO<sub>2</sub> permeability rates are closely linked to MAP film types. According to Robertson (2006), the oxygen transmission rates of PP, LDPE, and PVC films are 4,650; 7,750; and 155 cc m<sup>-2</sup> mil<sup>-1</sup> atm<sup>-1</sup> day<sup>-1</sup>. Meanwhile,

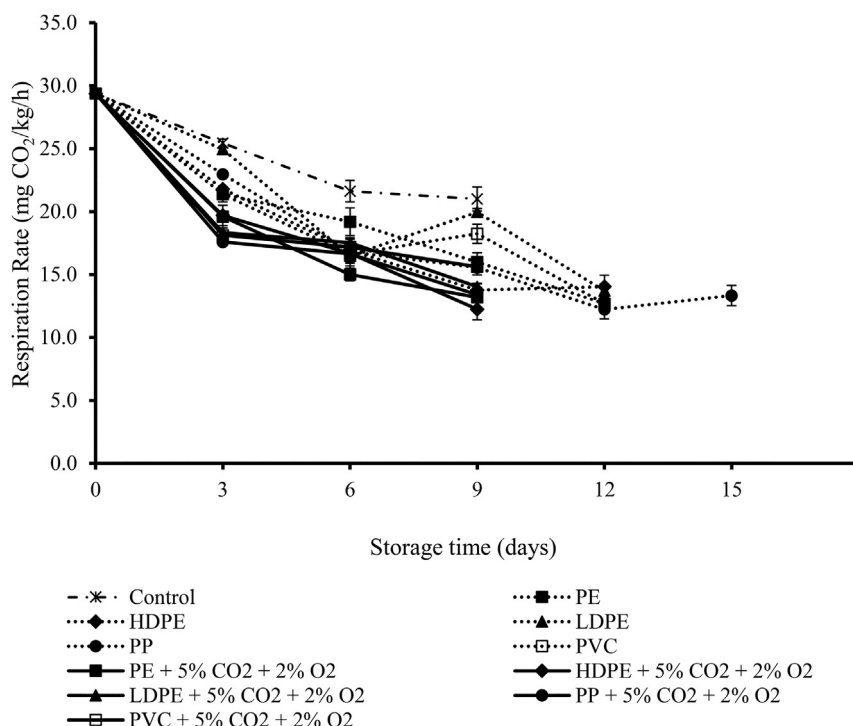
**Table 1.** Effects of passive and active MAP on storage life of cut *Dendrobium* Sonia orchid flowers.

MAP Type	Condition/Treatment	Storage life (Days)*
-	Control	7.67 <sup>d</sup> ± 0.57
Passive	PE	12.33 <sup>b</sup> ± 0.57
Passive	HDPE	12.33 <sup>b</sup> ± 0.57
Passive	LDPE	12.33 <sup>b</sup> ± 1.15
Passive	PP	15.66 <sup>a</sup> ± 1.15
Passive	PVC	13.00 <sup>b</sup> ± 0.57
Active	PE + 5% CO <sub>2</sub> + 2% O <sub>2</sub>	9.33 <sup>c</sup> ± 2.07
Active	HDPE + 5% CO <sub>2</sub> + 2% O <sub>2</sub>	9.33 <sup>c</sup> ± 0.97
Active	LDPE + 5% CO <sub>2</sub> + 2% O <sub>2</sub>	9.33 <sup>c</sup> ± 1.05
Active	PP + 5% CO <sub>2</sub> + 2% O <sub>2</sub>	9.33 <sup>c</sup> ± 1.15
Active	PVC + 5% CO <sub>2</sub> + 2% O <sub>2</sub>	9.33 <sup>c</sup> ± 1.15
Coefficient of variation (%)		7.55
Least significant difference (LSD)		1.95

\* The values are mean ± standard deviation; and different superscripts indicate statistical significance at 5 % ( $p < 0.05$ ).



**Figure 2.** Effects of active and passive MAP on the physical appearance of cut *Dendrobium Sonia* orchid flowers after 9 days: (a) under normal atmosphere condition, (b) in PP-film passive MAP, (c) in PP-film active MAP.



**Figure 3.** Effects of active and passive MAP on respiration rates of *Dendrobium* orchid flowers given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

the CO<sub>2</sub> permeability of PP, LDPE, and PVC films are 75.5; 131.6 and 1.64 cm cm<sup>-2</sup> sec<sup>-1</sup> mm Hg<sup>-1</sup> (Kumar et al., 2019; Robertson, 2006).

The composition of internal gases is closely related to plastic film type and thickness. By comparison, the O<sub>2</sub> permeability of LDPE film is higher than that of PP and PVC films (Reid et al., 1996; Kader, 2002). In comparison with air-stored flowers, those retained in PP-film passive MAP had lower water vapor transmission and longer shelf life (Singh et al., 2009). In addition, the orchid flowers wrapped in PP-film passive MAP had higher internal CO<sub>2</sub>, lower respiration rate, and lower ethylene production, vis-à-vis those stored in PP-film active MAP (Uthairatanakij et al., 2010; Poonsri, 2021).

### 3.5. Anthocyanin content

The anthocyanin content decreased with storage time. The anthocyanin content of the orchid flowers under normal atmosphere condition (control) and in active and passive MAP were statistically different (p < 0.05). Under the normal atmosphere condition, the anthocyanin content decreased from 8.54 (day 0) to 3.55 mg/100 g fresh weight (day 9). Besides, the vase life of orchid flowers at room temperature (25 °C) after 9 days of storage was shorter than seven days.

With PP-film passive MAP, the anthocyanin content decreased from 8.54 (day 0) to 4.22 mg/100 g fresh weight (day 15). In addition, the

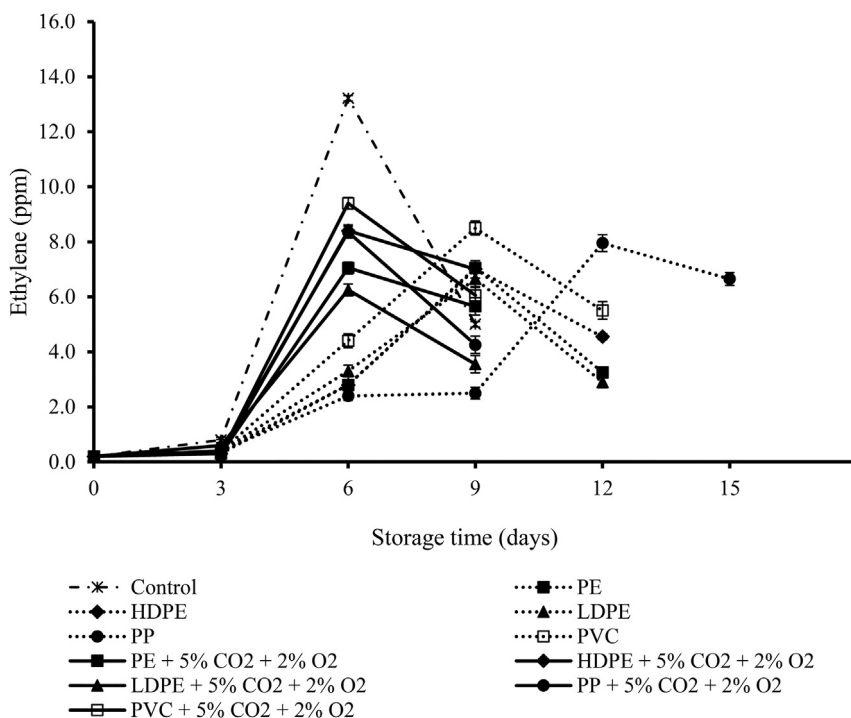


Figure 4. Effects of active and passive MAP on ethylene production of *Dendrobium* orchid flowers given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

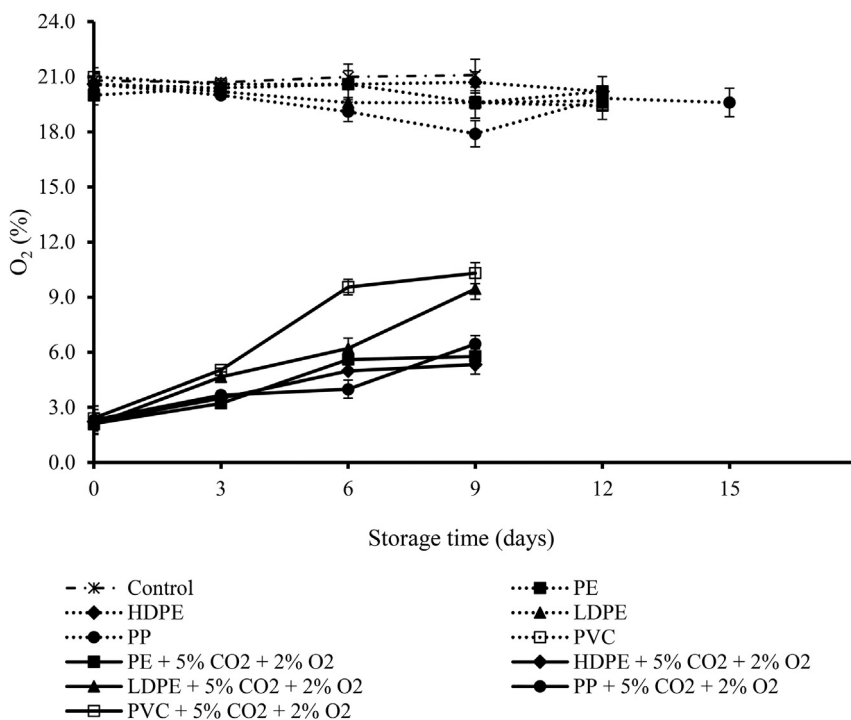
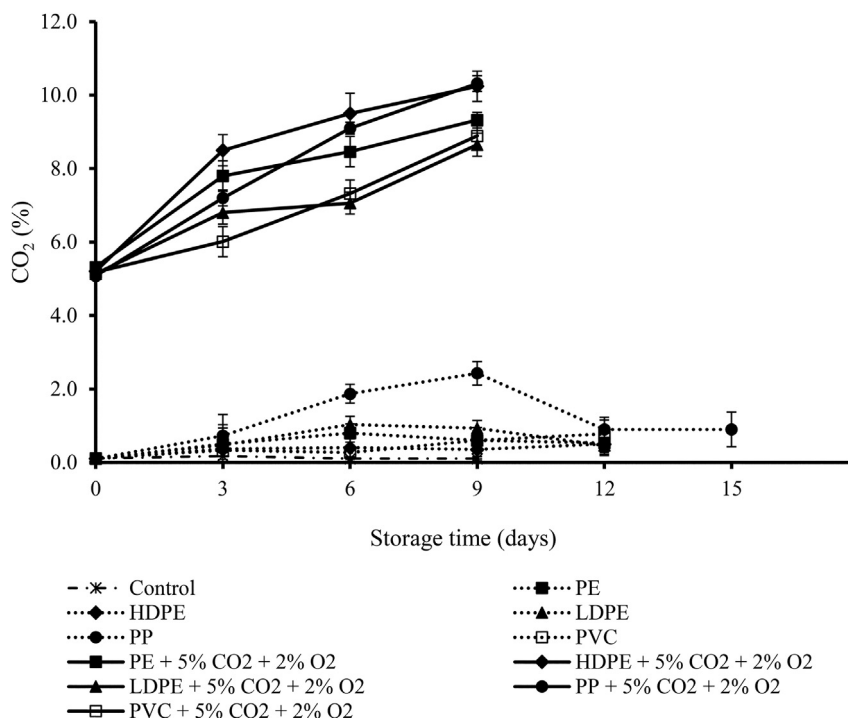


Figure 5. Effects of active and passive MAP on O<sub>2</sub> concentrations inside the packaging given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

orchid flowers after 15 days of storage lasted longer than 7 days since less than 50 % of the orchid flowers had wilted. With PP-film active MAP, the anthocyanin content decreased from 8.54 (day 0) to 5.09 mg/100 g fresh weight (day 9). The vase life of orchid flowers after 12 days of storage was less than seven days.

In Figure 7, the anthocyanin degradation of orchid flowers in active and passive MAP was slower than under the normal atmosphere condition (control). The slower anthocyanin degradation was attributed to lower injuries to the plant membranes stored in MAP (Hershkovitz et al.,



**Figure 6.** Effects of active and passive MAP on CO<sub>2</sub> concentrations inside the packaging given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

2005), resulting in lower loss of enzyme/substrate compartmentation and anthocyanin degradation (Jiang et al., 2004).

### 3.6. Protein degradation

In Figure 8, total protein in the orchid flowers under the normal atmosphere condition (control) rapidly decreased, unlike in active and passive MAP which gradually decreased. Under the normal atmosphere condition, total protein decreased from 343.9 (day 0) to 329.61 mg/g fresh weight (day 9). The results were consistent with the anthocyanin degradation.

With PP-film passive MAP, total protein gradually decreased from 341.9 (day 0) to 333.66 mg/g fresh weight (day 15). Meanwhile, the total protein of the orchid flowers retained in PP-film active MAP decreased from 343.9 (day 0) to 335.96 mg/g fresh weight after nine days. The protein degradation was closely related to the anthocyanin degradation and flower senescence (Poonsri, 2020).

Degradation of membrane-associated proteins induced flower senescence (Stephenson and Rubinstein, 1998; Hansel et al., 1993; Callis, 1995; Beers and Freeman, 1997; Ketsa and Rugkong, 2000). Poonsri (2017) also reported that protein degradation in cut *Dendrobium* orchid flowers caused senescence, wilting, and anthocyanin degradation.

The protein degradation in *Dendrobium* orchid flowers was also closely linked to ethylene synthesis and action as endogenous ethylene increased protease activity, which subsequently induced protein degradation (Callis, 1995; Hunter et al., 2004; Jones et al., 2005). The protein levels in the orchid flowers stored in active and passive MAP gradually decreased due to lower protease activity. Specifically, higher CO<sub>2</sub> and lower O<sub>2</sub> in MAP retarded ethylene action and protein degradation in the orchid flowers.

### 3.7. Electrolyte leakage

The electrolyte leakage of orchid flowers under normal atmosphere condition (control) increased rapidly, in comparison with those stored in active and passive MAP which gradually increased, as shown in Figure 9.

The electrolyte leakage under the normal atmosphere condition increased from 5.17 % (day 0) to 14.27 % (day 9), consistent with the anthocyanin and protein degradation.

With PP-film passive MAP, the electrolyte leakage gradually increased from 5.17 % (day 0) to 13.68 % (day 15), compared to under normal atmosphere condition. Meanwhile, with PP-film active MAP, the electrolyte leakage increased from 5.17 % (day 0) to 11.25 % (day 9). The findings were closely linked to the membrane-associated protein degradation, resulting in increased membrane permeability.

According to O'Donoghue et al. (2002b); Poonsri (2015), increased electrolyte leakage resulted in the senescence of orchid flowers due to higher cell loss as a result of increased activity of phospholipases, phosphatidic acid phosphatase and lipoxygenase (Hong et al., 2000). In addition, decreased membrane proteins also increased salute leakage (Celikel and van Doorn, 1995; Callis, 1995). Flower senescence was also closely related to membrane permeability (Wang, 1990). Increased membrane permeability led to petal senescence, loss of turgor, and visible wilting due to higher cellular electrolyte leakage (Mphahlele et al., 2020; Faragher et al., 1987). According to Leverentz et al. (2002), ethylene-induced leakage of solute from the cells resulted in petal senescence, loss of turgor, and wilting of *Alstroemeria* flowers.

Despite the widespread use to preserve the quality and prolong the storage life of fresh fruits and vegetables for exportation, the use of MAP for ornamental plants and flowers is limited. Apart from the results of this current research, existing evidence also demonstrates that MAP effectively extends the postharvest life of cut flowers (e.g., *Anthurium*, carnation, rose) and bulb crops (e.g., lily, tulip) (Andrew et al., 2009). As a result, the ornamentals industry and exporters should adopt the MAP technology to extend the shelf life of ornamentals, which would offer them with more trade opportunities to expand into farther foreign markets.

Furthermore, as one of the United Nations' 17 Sustainable Development Goals (SDGs), SDG 13 is about climate change whose aim is to take urgent action to combat climate change and its impacts. Since storing postharvest ornamentals in MAP lowers the CO<sub>2</sub> production inside the packaging and thus CO<sub>2</sub> to be released into the atmosphere, the MAP



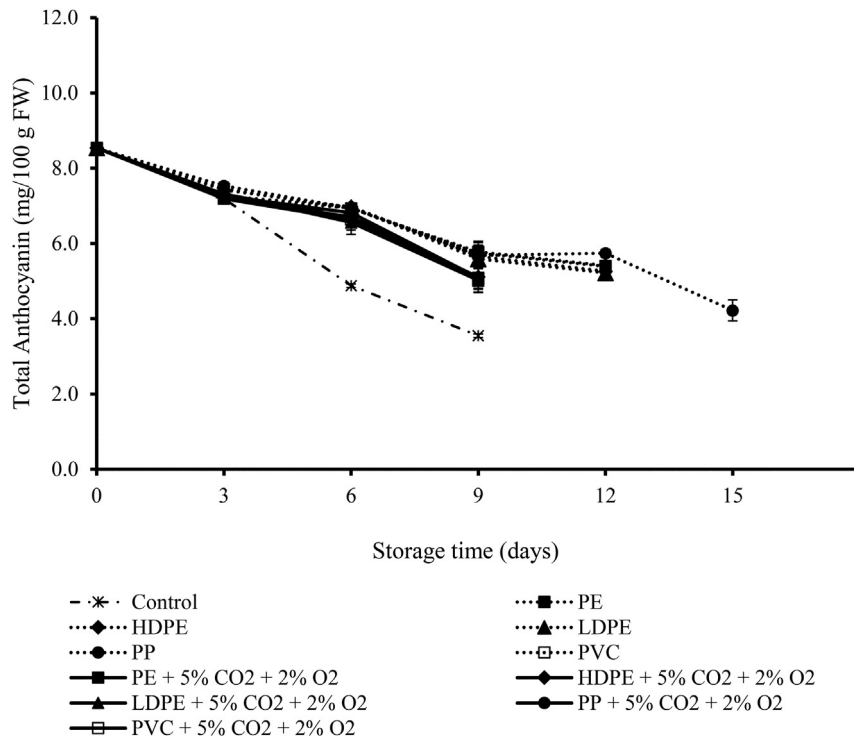


Figure 7. Effects of active and passive MAP on anthocyanin content given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

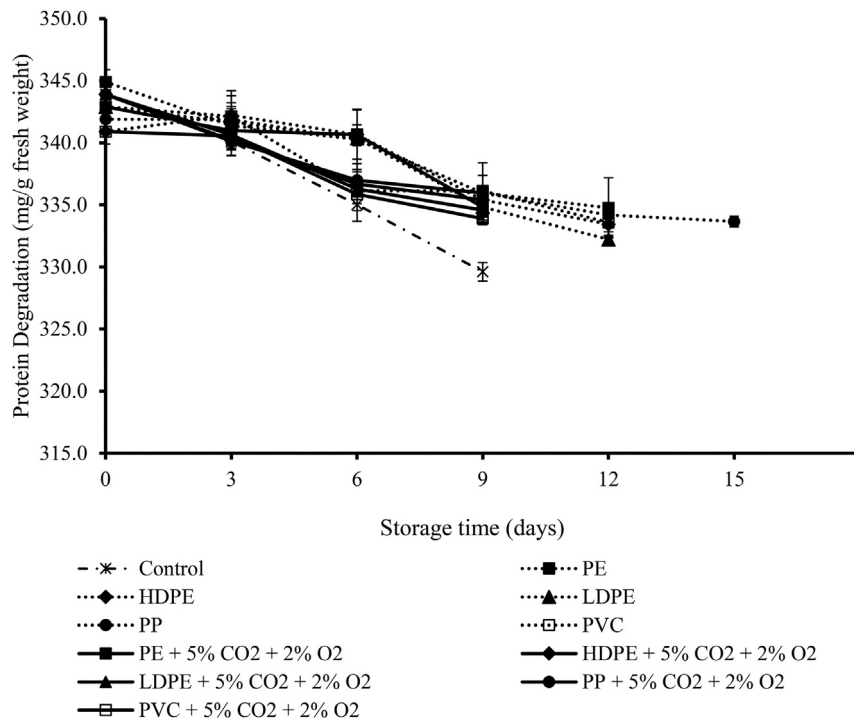
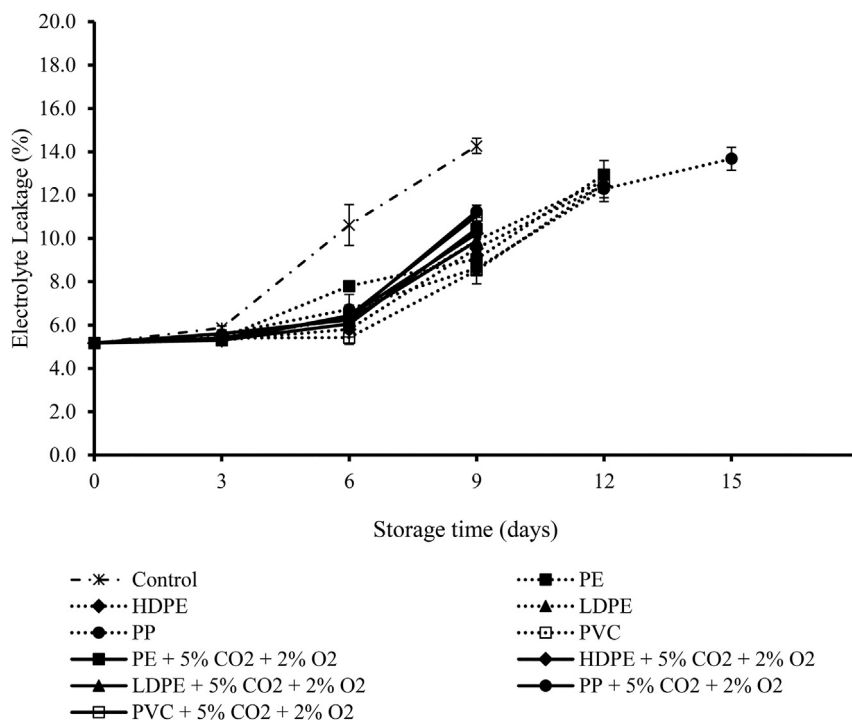


Figure 8. Effects of active and passive MAP on protein degradation given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.





**Figure 9.** Effects of active and passive MAP on electrolyte leakage given 13 °C storage temperature and 95 % RH. The error bars represent standard deviation.

technology could also be adopted as a means to combat climate change and mitigate its impacts, in addition to other climate actions.

#### 4. Conclusions

This research comparatively investigated the effects of active and passive MAP (i.e., PE, LDPE, HDPE, PP, and PVC films) on the biochemical properties of cut *Dendrobium Sonia* orchid flowers. The experimental results were also compared with those under normal atmosphere condition (control). Under active MAP conditions, the orchid flowers were wrapped with MAP film filled with 5 % CO<sub>2</sub> and 2 % O<sub>2</sub>. Meanwhile, under passive MAP conditions, the orchid flowers were stored in MAP without 5 % CO<sub>2</sub> and 2 % O<sub>2</sub>. The storage temperature and RH were 13 °C and 95 %, and all orchid flowers were retained in the dark. The results showed that the storage life of orchid flowers in passive MAP lasted between 12 – 15 days, with the flowers in PP-film passive MAP having the longest storage life (15.66 days). Meanwhile, the average storage life of orchid flowers in active MAP was 9.33 days. The storage life of orchid flowers under normal atmosphere condition (without MAP) was 7.67 days. In addition, MAP efficiently reduced the respiration rate, ethylene production, anthocyanin degradation, protein degradation, and electrolyte leakage, in comparison with without MAP. In essence, the MAP technology, particularly PP-film passive MAP, should be adopted by exporters of cut orchid flowers to extend the storage life and preserve the flower quality. The longer storage life would provide the exporters with greater business opportunities to sell to new and distant overseas markets.

#### Declarations

##### Author contribution statement

Warinthon Poonsri: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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##### Data availability statement

The data that has been used is confidential.

##### Declaration of interests statement

The authors declare no conflict of interest.

##### Additional information

No additional information is available for this paper.

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