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Standardization of packaging technique(s) for the distant marketing of *Rhynchostylis retusa*

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ABSTRACT

A postharvest experiment was conducted at the Department of Horticulture, AAU, Jorhat, to study the effect of different packaging technique(s) for the distant marketing of Foxtail orchid spikes. The seven treatments were as follows: T_1: CFB box (control); T_2: wrapping with 200 gauge polyethylene sheet + $KMnO_4$ sachet + CFB box; T_3: wrapping with butter paper + $KMnO_4$ sachet + CFB box; T_4: wrapping with Brown Paper + $KMnO_4$ sachet + CFB box; T_5: plastic vial containing 8-HQS (25 ppm) + wrapping with 200 gauge polyethylene sheet + CFB box; T_6: plastic vial containing 8-HQS (25 ppm) + wrapping with butter paper + CFB box; and T_7: plastic vial containing 8-HQS (25 ppm) + wrapping with Brown Paper + CFB box. Among these packaging treatments, T_5 resulted in a significantly longer vase life (7.33 days after 3 days of storage under ambient conditions). Postharvest parameters such as the PLW (6.58%), TSS (10.08%), MSI (78.28%), and wilting of the first floret (3.41 days) were found to be greatest in T_5. However, in terms of biochemical parameters, T_5 was similar to T6 in terms of carbohydrate content (63.49 and 62.08 mg/100 ml, respectively), protein content (15.23 and 15.18%, respectively) and anthocyanin content (6.37 and 6.30 mg/L, respectively).

Introduction

The Orchidaceae family is one of the largest angiosperm flowering plants, comprising more than 28,000 species, 5 subfamilies and 763 genera (Christenhusz and Byng, 2016), and accounts for nearly 8% of the total flowering plant species reported thus far (Willis, 2017). These plants are considered among the most diverse and evolved plants and are known for their rich flower diversity, *viz.* colors, form, textures, shape, size and fragrance (Peakall, 2007). Among them is *Rhynchostylis retusa* (L.) Blume, also known as the "Foxtail orchid", which belongs to the Vanda alliance. The flower has a long inflorescence that resembles a fluffy, tapering fox tail, and it consists of more than 100 pink-spotted white flowers that bloom in early spring. The Foxtail orchid is valued for its beautiful and distinctive blooms with an unusual range of colors as well as for its spicy aroma. The inflorescence is considered to be a symbol of love, fertility and merriment and has essential cultural significance in Assam, famously used as a hair-ornament worn by Assamese women during the

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Bihu folk dance on the onset of spring. Similarly, due to their distinctive flower shape, lengthy flowering duration, and opening around the Spring Festival, these plants are popular and well-concerned spring festival gifts in China. In addition to being a horticulturally important plant, it represents an enormous reservoir of biologically active chemical compounds that possess disease-preventive properties. Despite its importance and wide use, there is very little information available on *R. retusa,* and there are hardly any attempts to commercialize this orchid. In addition, the Foxtail orchid has great potential as a cut flower in the floricultural industry. From a marketing perspective, the quality of cut spikes is crucial both domestically and internationally. Postharvest management and value addition can increase the price of flowers. Postharvest handling involving packaging and proper packing has a significant role in determining the vase life and quality of the cut spike (Ghildiyal *et al.,* 2012). In addition, substandard packaging and storage practices can deteriorate the quality of products and shorten their life, which eventually decreases their market value. The quality of cut flowers is governed by internal mechanisms that include balancing water uptake, water loss, stem plugging, the respiration rate, the production of toxic substances such as ethylene and external factors such as environmental conditions and microbial attacks on cut ends. Packing aids in preserving quality and averts mechanical harm throughout the supply chain. Apart from that, it acts as a barrier between the conditions inside and outside the package, protecting the flowers from unfavorable outside conditions and enabling a microclimate to develop inside the package (Lavanya *et al.,* 2016).. Since flowers are delicate and have a relatively short shelf life, special care and appropriate packaging technologies are needed to keep them fresh. Moreover, the use of various wrapping materials results in a modified atmosphere (higher CO_2 and lower O_2) during storage, which lowers the metabolism of flowers and florets during storage and lowers ethylene production, which slows the consumption of carbohydrates as the respiratory substrate. A delay in floral senescence thus prolongs the life of cut flowers (Dilley, 2006). Moreover, the germicide used in the floral industry acts as an antimicrobial agent that can lead to increased water uptake. Foxtail orchid was once a traditional orchid, and despite its importance and wide use, very little information is available on the commercialization of this orchid. Therefore, standardization of packaging technique(s) for distant markets is highly important for the commercialization of crops, and practically no information is available on these crucial aspects. As a result, this research investigated the effects of different packaging technique(s) for the distant marketing of the Foxtail orchid.

Material and Methods

The experiment was conducted at the Department of Horticulture, Assam Agricultural University, Jorhat, during the years 2021-2022. Here, uniformly sized cut spikes of *Rhynchostylis retusa* with an average spike length of 35 cm were used in the packaging study. The basal ends of the flower stalks were cut, and each individual flower was packed according to the abovementioned treatments (Table 1). The wrapping materials were uniform in size (40 cm × 30 cm). The CFB box used in the experiment measured 45x20x15 cm with 2 vents, each with a 2.5 cm diameter. After the flowers were packed, the CFB boxes were sealed with cellophane tape and kept in the laboratory under ambient conditions for 3 days. The flowers were then unpacked, and observations were recorded for different parameters. Furthermore, (0.5 cm) stem ends were cut with sharp scissors, three spikes were placed in a conical flask containing 150 ml of distilled water, and the parameters were recorded after storage. Each treatment was replicated three times.

Physiological loss in weight: The physiological loss in weight was calculated as the difference between the initial weight (before storage) and the weight at the time of measurement (3 days after storage) and was expressed as a percentage.

Total soluble solids (TSS %): The total soluble solids were recorded in fresh sample florets (3 days after storage) using a Pal-1 pocket refractometer and are expressed as a percentage.

Membrane stability index (MSI %): Two samples (0.2 g) were taken (3 days after storage) and placed in 20 mL of double distilled water in two different 50 ml flasks. The first one (C1) was kept at 40°C for 30 min, while the second (C2) was kept at 100°C in a boiling water bath for 15 min. The electrical

Table 1: Different packaging treatments used in the experiment

Treatment	Materials
T_1	CFB box (Control)
T_2	Wrapping with 200-gauge polyethylene sheet +$KMnO_4$ sachet+ CFB box
T_3	Wrapping with Butter paper +$KMnO_4$ sachet+ CFB box
T_4	Wrapping with Brown Paper +$KMnO_4$ sachet+ CFB box
T_5	Plastic vial containing 8-HQS (25ppm) + Wrapping with 200-gauge polyethylene sheet+ CFB box
T_6	Plastic vial containing 8-HQS (25ppm) + Wrapping with Butter paper + CFB box
T_7	Plastic vial containing 8-HQS (25ppm) + Wrapping with Brown Paper + CFB box

***Plastic vial used in the T_5, T_6 and T_7 treatments containing 5 ml of solution of 8-HQS (25 ppm**

conductivity was measured with a conductivity meter. The leakage of ions was expressed as the membrane stability index according to the following formula:

MSI = [1 − (C1/C2)] × 100   Sairam *et al.* (1997)

Carbohydrate content (mg/100 ml): Flower samples from each treatment were taken after 3 days of storage for determination of carbohydrate content via the anthrone method (Hedge and Hofreiter, 1962).
Protein content (%): Flower samples from each treatment were taken after 3 days of storage, and the available N in the sample was estimated by Kjeldahl method as described by Jackson (1973). Thereafter, the protein content was calculated by using the formula % protein = 6.25 × % N.
Anthocyanin (mg/L): Flower samples from each treatment were taken after 3 days of storage, and the anthocyanin content was determined by using the method described by Yang *et al.* (2019).
Wilting of the first floret: After storing for 3 days, the flowers were removed from the packs. The stem ends were recut diagonally and placed in conical flasks containing distilled water, and wilting of the first floret was recorded from the end of the storage period to the time when the first floret started to wilt.
Vase life of flowers (days): After being stored for 3 days**,** the flowers were removed from the packs. The stem ends were recut diagonally and placed in conical flax flasks containing distilled water, and their life was noted by daily evaluation of the appearance of the flowers. Vase life was calculated from the end of the storage period to the time when more than 50% of the flowers were wilted**.**

Results and Discussion

As shown in Table 2, compared with those in all the treatments, including the control, the cut spikes packed in treatment T_5 resulted in significantly less physiological weight loss (6.58% after 3 days of storage under ambient conditions in the laboratory). Treatment T5 was subsequently administered, resulting in a 7.20% increase in the PLW. On the other hand, the highest PLW (12.87%) was recorded in the T_1 group. Pranuthi *et al*. (2018) also observed lower physiological weight loss in carnia plants treated with polyethylene than in control plants. Physiological weight loss may be associated with the rate of respiration of flowers and flower transpiration and leads to exhaustion of assimilates that have already accumulated in stem tissues. Weight loss is mainly the result of flower transpiration, where vapor-phase diffusion, which is driven by a gradient of water vapor pressures between the inside and outside of the produce, causes water to exit the flowers. Wrapping materials act as semipermeable barriers against O_2, CO_2, moisture and solute movement, thereby reducing respiration, water loss and oxidation rates (Robertson, 2006). The reduction in physiological weight loss in foxtail orchids during storage might be due to the wrapping of materials, i.e., the low water vapor transmission property of the polyethylene film, which prevents water loss and the

Table 2: Effect of packing technique(s) on the shelf life of *Rhynchostylis retusa* cut spikes

Treatment	PWL (%)	TSS (%)	MSI (%)	Carbohydrate content (mg/100 ml)	Protein content (%)	Anthocyanin (mg/L)	Wilting of first floret (days)	Vase life (days)
T_1	12.73	9.11	57.88	53.61	12.43	5.84	1.95	4.77
T_2	8.18	9.60	69.13	58.69	13.38	6.12	2.60	6.18
T_3	8.44	9.48	66.07	58.23	13.33	6.11	2.47	6.03
T_4	9.68	9.21	63.43	55.86	13.13	5.98	2.20	5.37
T_5	6.58	10.08	78.28	63.49	15.18	6.37	3.41	7.33
T_6	7.20	9.97	76.19	63.08	15.10	6.30	3.32	7.04
T_7	7.63	9.89	72.87	61.65	14.25	6.28	3.11	6.88
S.E (d) ±	0.27	0.02	0.90	0.84	0.16	0.03	0.04	0.13
C.D	0.59	0.03	1.96	1.81	0.33	0.06	0.07	0.28

***PWL- Physiological weight loss; TSS- Total soluble solids; MSI- Membrane stability index**

retention of relative humidity, and the germicidal action of 8-HQS, which triggers better water conductance through the stem. Significant differences were observed in terms of petal TSS values among the different treatments after three days of storage at ambient temperature. The cut spikes packed in treatment T_5 had the highest TSS content (10.08%), which was significantly different from that in all the other treatments. This treatment was followed by treatment with T_6 at 9.97%, while the lowest TSS value (9.11%) was recorded in treatment T_1. Foxtail orchid spikes in T_5 (plastic vial containing 8-HQS (25 ppm) + wrapping with 200-gauge polythene sheet+ CFB box) had the highest TSS values after 3 days of storage compared to those in the other treatment groups due to decreased physiological weight loss, decreased water loss and low sugar stress; these differences might be due to the greater amount of stored food in the floral organs. Moreover, according to (Makwana *et al.,* 2015), a high water balance, high water uptake and high fresh weight of stems and petals generate high petal TSS, and a correlation between TSS and water uptake has been indicated in rose plants. Our results are in accordance with those of Patel *et al.* (2016), who observed higher TSS values in wrapped Gerbera cut flowers than in unwrapped flowers. Furthermore, 8-HQS must have helped to maintain an adequate water balance as a result of increased water uptake and inhibition of bacterial growth in cut stems. The MSI was significantly influenced by the different treatments. The highest MSI (78.28%) was recorded in treatment T_5. This treatment was followed by treatment T_6, for which the average mortality rate was 76.19%. However, the lowest MSI (57.88%) was recorded in the T_1 subgroup, in which the spikes were simply placed in the CFB box. Packaging of the Foxtail orchid spikes using polyethylene in treatment T_5 created equilibrium of modified atmosphere (EMA) with high CO_2 and low O_2, resulting in less ethylene production. Our findings are also in line with Khongwir *et al.* (2019), who reported higher MSI in polyethylene-wrapped flowers in Tuberose plants. Moreover, decreased ethylene production causes a reduction in membrane permeability and ultimately increases the membrane stability of foxtail orchid petals. Furthermore, adequate water balance water levels due to increased water uptake and TSS levels in petal cells of PE-packaged cut flowers retained biomembrane fluidity and membrane permeability, which contributed to the high MSI of petal tissue. The data presented in Table 2 reveal that the T_5 treatment had the highest carbohydrate content, followed by the T_6 treatment (with averages of 63.49 mg/100 mg and 63.08 mg/100 ml, respectively). This clearly showed that no significant difference in carbohydrate content was found between cut spikes wrapped with 200-gauge polyethylene combined with dipping the cut end of the spikes in 25 ppm 8-HQS solution and cut spikes wrapped with butter paper combined with dipping the cut end of the spikes in 25 ppm 8-HQS. On the other hand, the lowest carbohydrate content (53.46 mg/100 ml) was recorded in the T_1 treatment, in which the spikes were simply placed in the CFB box. A longer vase life may be attributed to the higher concentration of soluble carbohydrates in petals (Nabigol *et al.,* 2014). Cutflowers have a limited carbon source, but sucrose decomposition and glucose consumption decrease carbohydrate content (Norikoshi *et al.,* 2016). The greater carbohydrate content in the wrapped cut foxtail orchid spikes than in the nonwrapped spikes might be due to the greater amount of stored food in the floral organs. This is because wrapping cut stems with polyethylene results in a beneficial equilibrium of modified atmosphere (EMA), which leads to closer stomata. These findings are supported by Kumari *et al.* (2017), who performed similar research on chrysanthemum flowers. Furthermore, the 8-HQS provided in vials as wet packaging must have maximized water uptake and maintained the water balance in the flowers. Table 2 clearly shows that T_5 was on par with T_6, with average protein contents of 15.23% and 15.18%, respectively. However, the lowest protein content was recorded in T_1, with an average value of 12.60%. Wrapping of cut stems created higher CO_2 and lower O_2 conditions, which helped to store more food and retarded ethylene action as well as protein degradation in the orchid cut spikes. Poonsri (2020) reported that using HDPE decreased protein degradation in cut flowers of *Dendrobium.* Additionally, the use of 8-HQS in a holding solution for wet packaging for long-distance transport was found to help delay flower weight loss and reduce dry weight, thus delaying protein degradation. In our study, the lower protein content in unwrapped

Rhynchostylis retusa flowers (T_1) might also be correlated with ethylene synthesis. The anthocyanin content was significantly influenced by the different treatments. Among the treatments compared, the highest anthocyanin content (6.37 mg/L) was recorded in the T_5 treatment. This was followed by treatment (T_6), in which the average anthocyanin content was 6.30 mg/L. Moreover, the lowest anthocyanin content (5.84 mg/L) was recorded in the T_1 treatment. Packaging the cut spike reduces damage to the membranes and thus decreases the loss of compartmentation between enzymes and their substrates, thereby delaying anthocyanin degradation (Jiang *et al.,* 2004). Similar results were reported by Poonsri (2021), who reported that the use of polyethylene to wrap Dendrobium cut flowers increased the anthocyanin content compared to that in nonwrapped flowers. In addition, the continuous availability of 8-HQS in vials must help to inhibit bacterial growth in cut stems and maintain water uptake, thus preserving the fresh weight loss of the cut flowers and leading to slower anthocyanin degradation (Dineshbabu *et al.,*2002). Here, the spikes in the T_5 treatment showed delayed senescence, i.e., it took as long as 3.41 days for the first floret to wilt. T_5 was found to be significantly superior to all the other treatments in terms of this parameter. On the other hand, in the T_1 treatment, the first floret first wilted on the spikes within 1.95 days after removal from the CFB box. When the spikes were packed with polyethylene and placed in a CFB box, there was less moisture loss and high relative humidity inside. The turgidity of the spikes increased due to the conservation of moisture. Because of this, spikes remain fresh for longer periods of time after storage, thus delaying wilting of florets in foxtail orchids. Taken together, our results coincide with those of Sharma *et al.* (2008), who reported delayed wilting of the *Asiatic hybrid lily* cultivar 'Apeldoorn' when packed in polyethylene. Furthermore, 8-HQS in holding solution acted as an antimicrobial agent during storage and reduced stem plugging in foxtail orchids. This prevented the growth of microorganisms in the xylem, thus increasing water uptake by the flower stem, which ultimately delayed the wilting of the florets. The longest vase life (7.33 days) of the foxtail orchid spikes (more than 50% fading of florets after 3 days of storage) was recorded in treatment T_5. On the other hand, the shortest vase life (4.71 days) of foxtail orchid spikes was recorded in T_1. The vase life was extended by 2.56 days compared to that of the control. Wrapping of cut flower spikes of the foxtail orchid in polyethylene promoted the beneficial equilibrium of modified atmosphere closure of stomata and a reduction in the respiratory loss of carbohydrates in addition to transpiration loss of water from cut spikes (Zeltser *et al.,* 2001), causing minimal cell damage during storage and maintaining normal cell conditions after storage. Therefore, there was greater vase solution uptake by the cut spikes after storage, and when more vase solution was absorbed by the cut spikes, the vase life increased. Similarly, according to Kumari *et al.* (2017), the use of polyethylene extended the life of chrysanthemum plants by 5.48 days. Moreover, 8-HQS must improve the stem hydraulic conductance of foxtail orchids while preserving the fresh weight of the cut flowers, thus improving the vase life of the cut flower stems. Dineshbabu *et al.* (2002) previously reported that using 8-HQS as a holding solution increases the freshness of Dendrobium cut flowers.

Conclusion

It may be concluded that the packaging materials and chemicals used in this study improved the quality of the cut foxtail orchid flowers. Among all the packaging treatments compared, plastic vials containing 8-HQS (25 ppm) + wrapped with 200-gauge polythene sheets + CFB boxes resulted in a significantly longer vase life (7.33 days after 3 days of storage in ambient conditions). Postharvest parameters such as PLW (6.85%), TSS (10.08%), MSI (78.28%), and wilting of the first floret (3.41 days) were found to be superior in plastic vials containing 8-HQS (25 ppm) + wrapping with a 200-gauge polythene sheet+ a CFB box. The above treatment was performed on par with plastic vials containing 8-HQS (25 ppm) + wrapping with butter paper + CFB in terms of carbohydrate content (63.49 and 63.08 mg/100 ml, respectively), protein content (15.32 and 15.18%, respectively), and anthocyanin content (6.37 and 6.30 mg/L). Therefore, for long-distance transport of cut spikes, it is recommended to wrap the cut spikes with 200-gauge polyethylene combined with dipping the cut ends of the spikes in plastic vials containing 8-HQS (25 ppm) and placing them in a CFB box with a 2.5 cm vent on both sides.

Conflict of interest

The authors declare that they have no conflicts of interest.

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