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EFFECT OF PULSING WITH GROWTH REGULATORS ON SENESCENCE OF THE DETACHED COLD-STORED LEAVES OF *Zantedeschia aethiopica* Spr. AND *Hosta* 'Undulata Erromena'

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Abstract. Cut leaves of *Zantedeschia aethiopica* Spr. and *Hosta* 'Undulata Erromena' are widely used as the florists' green. Effect of pulsing leaves with growth regulators and a storage method (dry or wet, in light or darkness, at 5°C) on postharvest longevity and chlorophyll content was investigated. Leaf vase life after 3 days storage was evaluated at 20°C and 12 h light/12 h night. Results confirmed that a dry and dark storage of *Zantedeschia* leaves negatively affected their decorative values while for *Hosta* foliage a dry storage was improper both in darkness and in light. Negative effects of storage conditions could – at least partly – be alleviated by a 24 h puls conditioning with gibberellic acid or benzyladenine for *Zantedeschia* and *Hosta*, respectively. In *Zantedeschia* the pulsed leaves cold stored in darkness had their vase life seven-fold that of non pulsed leaves. In *Hosta* pulsing leaves with BA prior to storage increased their vase life six times relative to untreated leaves, however, the cytokinin did not completely overcome a negative effect of dry storage. Positive action of both growth regulators was reflected in the leaf higher chlorophyll contents at all sampling dates.

Key words: *Zantedeschia aethiopica*, *Hosta* 'Undulata Erromena', cold storage, growth regulators, senescence, chlorophyll content, florists' green

INTRODUCTION

Zantedeschia aethiopica and *Hosta* 'Undulata Erromena' are plants of commercial importance providing the florists' green widely used in cut flower arrangements. As their importance on the international cut flower markets grows it seems indispensable to elaborate the most efficient handling methods on every step of the market chain from a grower through a wholesaler and finely to a consumer. Growth regulators such as gibberellic acid and cytokinins improve keeping qualities of cut leaves, however,

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a foliage response to growth regulators varies from species to species [Skutnik et al. 2003]. Postharvest longevity of cut *Hosta* leaves can be extended by benzyladenine applied as a dip (1 mmol·dm^{-3}) or the 24 h puls conditioning (0.10 mmol·dm^{-3}) [Skutnik and Łukaszewska 2001] while vase life of *Zantedeschia* leaves can be prolonged by gibberellic acid (0.25 mmol·dm^{-3}) [Skutnik et al. 2001].

Postharvest quality of cut foliage can also be maintained by appropriate storage methods [Rudnicki and Nowak 1992]. Earlier studies on detached leaves of *Zantedeschia* and *Hosta* showed that even a short dry storage in darkness, often practiced by growers or wholesalers, significantly shortens the foliage display life [Rabiza-Świder and Skutnik 2005]. It seems therefore necessary to find an appropriate storage method and/or treatments which could alleviate negative effects of storage and delay senescence in leaves used as the florists' green. An application of growth regulators seems a promising approach results of which are presented in this paper.

MATERIAL AND METHODS

Detached leaves of *Zantedeschia aethiopica* Spr. and *Hosta* 'Undulata Erromena' harvested from plants grown in the didactic collections of the Department of Ornamental Plants were used in the experiments. Mature leaves were harvested in the morning and immediately transported to the laboratory. Leaves were either directly placed into vases with distilled water or pulsed 24 hours with distilled water, 0.25 mmol·dm^{-3} GA_3 or 0.10 mmol·dm^{-3} BA, respectively and placed in phytotrone with 20°C, RH 60%, irradiation 35 μmol·m^{-2}·s^{-1}, 12 hours light and 12 hours night. After the treatment leaves were stored 3 days at 5°C as follows: in light/wet, in light/dry, in darkness/wet and in darkness/dry. Wet stored leaves were placed into vases with distilled water and covered with transparent polyethylene perforated foil (storage in light) or with black foil (storage in darkness). Dry stored leaves were placed in a sleeve of either transparent or black foil. Irradiation during storage period was 3.5 μmol·m^{-2}·s^{-1}. After storage leaves were placed in vases with distilled water and transferred to a phytotrone. Non stored leaves placed in the phytotrone immediately after harvest served as a control treatment.

Effect of a storage method on leaf vase life and chlorophyll contents was determined. Vase life was a number of days till leaves lost their decorative values, i.e. when 30% of a leaf blade showed such symptoms as discoloration, yellowing, blade edge browning or drying. Ten leaves were evaluated in each treatment, serving as individual replicates. Separate batches of leaves were treated as above and served as sampling material for chlorophyll analyses and dry weight determination. Chlorophyll was extracted in DMF and determined according to Moran and Porath [1980] in modification of Inskeep and Bloom [1985]. Results were calculated as mg chlorophyll a + b per gram DW.

Results were statistically evaluated by ANOVA 2 or ANOVA 3. Means were compared using the Duncan's test at probability level $\alpha = 0.05$.

RESULTS AND DISCUSSION

According to Rudnicki and Nowak [1992] cut flowers and leaves should be cooled immediately after harvest at a temperature optimal for a given species. Low temperature decreases respiration rate and delays respiratory substrate depletion, lowers ethylene production, diminishes water losses and inhibits pathogen development [Łukaszewska and Skutnik 2003]. Cold storage at 1°C delayed chlorophyll degradation in spinach [Yamauchi and Watada 1991]. A rapid cooling of cut Irish bells (*Molucella laevis*) to 6–10°C decreased both the endogenous ethylene production and plant sensitivity to exogenous C_2H_4 [Skutnik 1998]. Earlier studies on *Z. aethiopica* and *Hosta* 'Undulata Erromena' showed that though keeping leaves 3 days at 5°C did not shorten their vase life afterwards, a method of cold storage could significantly decrease foliage postharvest longevity [Rabiza-Świder and Skutnik 2005]. Storing *Hosta* leaves dry, both in light and in darkness, hastened their senescence. For *Zantedeschia* darkness seemed to be more detrimental, especially when combined with a dry storage. In this work such a storage method decreased *Zantedeschia* vase life fourfold relative to non stored foliage (tab. 1).

Endogenous hormones control plant senescence, cytokinins and gibberellins retarding degradative processes [Nooden et al. 1997]. Exogenous gibberellins delay leaf yellowing in *Lilium longiflorum* [Han 2000], alstroemeria [Kappers et al. 1998], ivy [Horton and Bourguoin 1992], oat [Dhindsa et al. 1982] and *Hippeastrum hybridum* [Skutnik and Łukaszewska 2001]. Also in *Zantedeschia* GA_3 extends postharvest longevity [Skutnik et al. 2001], delaying processes symptomatic for senescence [Skutnik et al. 2003], like chlorophyll degradation [Skutnik et al. 2001, 2004] and proteolysis [Rabiza-Świder et al. 2003, 2004b] as well as proline and ammonia accumulation [Rabiza-Świder et al 2004a]. In this work an effect of puls conditioning with GA_3 was applied to *Zantedeschia* leaves in hope to improve their display characteristics after cold storage (tab. 1). Indeed, the treatment completely counteracted the effect of the 3 day storage at 5°C. The pulsed leaves from all the storage treatments had their vase life 33–36 days – on the average twice of that in leaves untreated before storage. Especially spectacular was the GA_3 action in the dry and dark stored leaves: those pulsed with 0.25 $mmol(GA_3)\cdot dm^{-3}$ before storage had their vase life almost sevenfold of that observed in the non pulsed leaves stored in the same conditions. The positive effect of GA_3 was confirmed by results of the chlorophyll a + b analyses in *Zantedeschia* leaves (tab. 2). Regardless the storage method leaves pulsed with gibberellic acid had on the average 36% more chlorophyll than had leaves pulsed with water. On the last sampling date the chlorophyll content in the GA_3-treated leaves dropped only by 42% as compared to the freshly harvested leaves, while in non treated leaves the chlorophyll content decreased 62%. Even in leaves subjected to dry storage in darkness which is the least favorable for *Zantedeschia* foliage the GA_3 puls retarded chlorophyll degradation maintaining the pigment content at 50% of the initial level while in the non pulsed leaves it fell to 27% (tab. 2).

Another group of hormones retarding senescence are cytokinins. During leaf senescence levels of endogenous cytokinins fall [Gan and Amasino 1997]. Such hormone losses are even more pronounced in leaves detached from a root system. To compensate

Table 1. Effect of storage method at 5°C and 24 hrs pulse conditioning with 0.25 mmol $(GA_3) \cdot dm^{-3}$ on vase life of *Zantedeschia aethiopica* leaves
Tabela 1. Wpływ sposobu przechowywania w 5°C i 24-godzinnego kondycjonowania liści cantedeskii etiopskiej w 0,25 mmo l $(GA_3) \cdot dm^{-3}$ na ich pozbiorczą trwałość

Storage method Sposób przechowywania	Vase life, days Trwałość, dni		Mean for a storage method Średnia dla sposobu przechowywania $LSD_{0.05}$ = 3.99; $NIR_{0,05}$ = 3,99
	H_2O	GA_3	
Without storage (control); Bez przechowywania (kontrola)	20.4	36.1	28.3 c[1]
Light / wet; Na świetle /na mokro	20.7	35.6	28.1 c
Light / dry; Na świetle /na sucho	16.7	33.6	25.1 bc
Darkness / wet; W ciemności /na mokro	14.4	32.6	23.5 b
Darkness / dry; W ciemności /na sucho	4.9	33.7	19.3 a
Mean for a treatment $LSD_{0.05}$ = 2.52 Średnia dla traktowania $NIR_{0,05}$ = 2,52	15.4 a	34.3 b	

[1] Means followed by the same letter do not differ significantly at α = 0.05 (Duncan's test). To compare the means within the table $LSD_{0.05}(A/B)$ = 5.64
[1] Średnie oznaczone tą samą literą nie różnią się istotnie przy α = 0,05 (test Duncana). Dla porównania pozostałych średnich $NIR_{0,05}(A/B)$ = 5,64

Table 2. Effect of storage method at 5°C and 24hrs pulse conditioning with 0.25 mmol $(GA_3)\cdot dm^{-3}$ on total chlorophyll content ($mg\cdot g^{-1}$ DW) in cut leaves of *Zantedeschia aethiopica.* Chlorophyll content immediately after harvest: 24.9 $mg\cdot g^{-1}$ DW

Tabela 2. Wpływ sposobu przechowywania w 5°C i 24-godzinnego kondycjonowania w 0,25 mmol $(GA_3)\cdot dm^{-3}$ na zmiany zawartości chlorofilu *a+b* ($mg\cdot g^{-1}$s.m.) w ciętych liściach cantedeskii etiopskiej. Zawartość chlorofilu a+b bezpośrednio po zbiorze: 24,9 $mg\cdot g^{-1}$s.m.

Treatment Traktowanie	Storage method Sposób przechowywania	Chlorophyll content after days: Zawartość chlorofilu *a+b* w dniu			$LSD_{0.05}$(A/B) = 0.17 $NIR_{0,05}$(A/B) = 0,17	Mean for a treatment $LSD_{0.05}$ = 0.08 Średnia dla traktowania $NIR_{0,05}$ = 0,08
		5	18	27		
H_2O	Without storage; Bez przechowywania	20.6	12.8	11.8	15.1 d[1]	13.1 a
	Light / wet; Na świetle /na mokro	20.9	12.9	11.8	15.2 d	
	Light / dry; Na świetle /na sucho	18.6	11.9	7.8	12.8 c	
	Darkness / wet; W ciemności /na mokro	19.1	9.8	8.8	12.6 b	
	Darkness / dry; W ciemności /na sucho	16.1	7.2	6.8	10.1 a	
$LSD_{0.05}$(A/C) = 0.13; $NIR_{0,05}$(A/C) = 0,13		19.1 e	10.9 b	9.4 a		
GA_3 24h	Without storage; Bez przechowywania	22.9	19.5	17.0	19.8 h	17.8 b
	Light / wet; Na świetle /na mokro	22.9	19.5	17.1	19.8 h	
	Light / dry; Na świetle /na sucho	22.1	14.6	13.0	16.6 f	
	Darkness / wet; W ciemności /na mokro	22.0	15.8	12.9	16.9 g	
	Darkness / dry; W ciemności /na sucho	21.2	14.2	12.4	15.9 e	
$LSD_{0.05}$(A/C) = 0.13; $NIR_{0,05}$(A/C) = 0,13		22.2 f	16.7 d	14.5 c		

Storage method Sposób przechowywania	Chlorophyll content after days Zawartość chlorofilu a+b w dniu:			Mean for a storage method Średnia dla sposobu przechowywania $LSD_{0.05}$ = 0.12; $NIR_{0,05}$ = 0,12
	5	18	27	
Without storage; Bez przechowywania	21.7 k	16.2 g	14.4 f	17.4 c
Light / wet; Na świetle /na mokro	21.9 k	16.2 g	14.4 f	17.5 c
Light / dry; Na świetle /na sucho	20.3 i	13.2 e	10.4 b	14.7 b
Darkness / wet; W ciemności /na mokro	20.5 j	12.8 d	10.8 c	14.7 b
Darkness / dry; W ciemności /na sucho	18.7 h	10.7 c	9.6 a	13.0 a
Mean for a term $LSD_{0.05}$ = 0.10 Średnia dla terminu $NIR_{0,05}$ = 0,10	20.6 c	13.8 b	11.9 a	

[1] Footnotes as in table 1. To compare the means within the table $LSD_{0.05}$(ABC) = 0.29

[1] Opis jak w tabeli 1. Dla porównania pozostałych średnich $NIR_{0,05}$(ABC) = 0,29

Table 3. Effect of storage method at 5°C and 24hrs pulse conditioning with 0.10 mmol (BA)·dm^{-3} on vase life of *Hosta* 'Undulata Erromena' leaves
Tabela 3. Wpływ sposobu przechowywania w 5°C i 24-godzinnego kondycjonowania liści funkii w 0,10 mmol (BA)·dm^{-3} na ich pozbiorczą trwałość

Storage method Sposób przechowywania	Vase life (days) Trwałość (dni)		Mean for a storage method Średnia dla sposobu przechowywania $LSD_{0.05}$ = 5.13; $NIR_{0,05}$ = 5,13
	H_2O	BA	
Without storage; Bez przechowywania	9.4	60.7	35.1 b[1]
Light / wet; Na świetle /na mokro	9.0	60.3	34.7 b
Light / dry; Na świetle /na sucho	6.0	38.4	22.2 a
Darkness / wet; W ciemności /na mokro	8.8	54.4	31.6 b
Darkness / dry; W ciemności /na sucho	7.6	45.3	26.4 a
Mean for a treatment $LSD_{0.05}$ = 3.25 Średnia dla traktowania $NIR_{0,05}$ = 3,25	8.2 a	51.8 b	

[1] Footnotes as in table 1. To compare the means within the table $LSD_{0.05}$(A/B) = 7.26
[1] Opis jak w tabeli 1. Dla porównania pozostałych średnich $NIR_{0,05}$(A/B) = 7,26

Table 4. Effect of storage method at 5°C and 24hrs pulse conditioning with 0.10 mmol (BA)·dm^{-3} on total chlorophyll content (mg·g^{-1} DW) in cut leaves of *Hosta* 'Undulata Erromena'. Chlorophyll content immediately after harvest: 16.8 mg·g^{-1} DW

Tabela 4. Wpływ sposobu przechowywania w 5°C i 24 godzinnego kondycjonowania w 0,10 mmol (BA)·dm^{-3} na zmiany zawartości chlorofilu *a+b* (mg·g^{-1}s.m.) w ciętych liściach funkii. Zawartość chlorofilu a+b bezpośrednio po zbiorze: 16,8 mg·g^{-1}s.m.

Treatment Traktowanie	Storage method Sposób przechowywania	Chlorophyll content after days: Zawartość chlorofilu *a+b* w dniu: 2	13	60	$LSD_{0.05}$(A/B) = 0.38 $NIR_{0,05}$(A/B) = 0,38	Mean for a treatment $LSD_{0.05}$ = 0.17 Średnia dla traktowania $NIR_{0,05}$ = 0,17
H_2O	Without storage; Bez przechowywania	14.5	10.3	0.2	8.3 b[1]	7.7 a
	Light / wet; Na świetle /na mokro	14.8	10.3	0.2	8.4 b	
	Light / dry; Na świetle /na sucho	13.4	6.4	0.2	6.7 a	
	Darkness / wet; W ciemności /na mokro	14.5	9.8	0.2	8.2 b	
	Darkness / dry; W ciemności /na sucho	14.5	5.3	0.2	6.7 a	
$LSD_{0.05}$(A/C) = 0.29; $NIR_{0,05}$(A/C) = 0,29		14.3 e	8.4 b	0.2 a		
BA 24h	Without storage; Bez przechowywania	16.2	14.5	12.7	14.5 e	13.6 b
	Light / wet; Na świetle /na mokro	16.2	14.7	12.7	14.5 e	
	Light / dry; Na świetle /na sucho	15.4	12.8	10.1	12.8 c	
	Darkness / wet; W ciemności /na mokro	16.0	13.2	12.5	13.9 d	
	Darkness / dry; W ciemności /na sucho	15.0	12.8	9.9	12.6 c	
$LSD_{0.05}$(A/C) = 0.29; $NIR_{0,05}$(A/C) = 0,29		15.8 f	13.6 d	11.6 c		

Storage method Sposób przechowywania	Chlorophyll content after days: Zawartość chlorofilu a+b w dniu: 2	13	60	Mean for a storage method Średnia dla sposobu przechowywania $LSD_{0.05}$ = 0.28; $NIR_{0,05}$ = 0,28
Without storage; Bez przechowywania	15.3 h	12.4 f	6.5 b	11.4 c
Light / wet; Na świetle /na mokro	15.5 h	12.5 f	6.5 b	11.5 c
Light / dry; Na świetle /na sucho	14.4 g	9.6 d	5.2 a	9.7 a
Darkness / wet; W ciemności /na mokro	15.3 h	11.5 e	6.4 b	11.0 b
Darkness / dry; W ciemności /na sucho	14.8 g	9.0 c	5.1 a	9.6 a
Mean for a term $LSD_{0.05}$ = 0.21 Średnia dla terminu $NIR_{0,05}$ = 0,21	15.1 c	11.0 b	5.9 a	

[1] Footnotes as in table 1. To compare the means within the table $LSD_{0.05}$(ABC) = 0.65

[1] Opis jak w tabeli 1. Dla porównania pozostałych średnich $NIR_{0.05}$(ABC) = 0,65

this shortage of endogenous hormone the exogenous cytokinins are delivered to cut leaves in order to delay their yellowing what is crucial for the florists' green. Skutnik and Łukaszewska [2001] showed that conditioning or dipping leaves in benzyladenine solutions extends postharvest longevity of *Asparagus densiflorus* 'Sprengerii' and *Hosta*. Antisenescence action of cytokinins is due – among others – to delaying chlorophyll degradation [Skutnik et al. 1999] and decreasing proteolysis [Rabiza-Świder et al. 2003]. Also in this work applying BA to *Hosta* leaves before storage improved their vase life: regardless the storage method it was six fold of that found in non pulsed stored leaves (tab. 3). The BA treated leaves had also more chlorophyll a + b (tab. 4): average content was 13.6 $mg \cdot g^{-1}$ DW as compared to 7.7 $mg \cdot g^{-1}$ DW in untreated leaves. Only 60 days after harvest the pigment content in the BA conditioned leaves dropped by 31% while in the untreated leaves it fell to half of the initial level already on 13th days of vase life.

Both vase life (tab. 3) and chlorophyll content (tab. 4) were affected by a storage method. Earlier work showed that dry storage can diminish display life of cut *Hosta* leaves [Rabiza-Świder and Skutnik 2005] what was confirmed here (tab. 3). Even the BA pulsed leaves when stored dry (both in light and in darkness) had their vase life shortened by around 25–37% (i.e. almost 19 days) as compared to leaves stored in water and those non stored. Also chlorophyll degradation proceeded faster in such leaves (tab. 4). Already on the 13th day the chlorophyll content in dry stored *Hosta* leaves (both in light and darkness) was by 12% lower than in unstored leaves or those kept in water during storage. On the 60th day the dry stored leaves had 20–22% less chlorophyll than those wet stored. However, it is worthy to note that BA application resulted in the six fold increase in leaf vase life relative to the BA untreated leaves and stored in the same conditions (tab. 3).

CONCLUSIONS

1. Puls conditioning cut leaves with growth regulators before a storage extends their subsequent display life. Gibberellic acid counteracts a negative effect of darkness during 3 day storage at 5°C of *Zantedeschia* leaves while benzyladenine increases vase life of the stored *Hosta* leaves though is not able to neutralize completely the negative effect of dry storage.

2. Positive effects of both growth regulators are related to delaying chlorophyll degradation in leaves of both species under study.

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WPŁYW KONDYCJONOWANIA REGULATORAMI WZROSTU NA STARZENIE SIĘ CIĘTYCH LIŚCI CANTEDESKII (*Zantedeschia aethiopica* Spr.) I FUNKII (*Hosta* 'Undulata Erromena') PRZECHOWYWANYCH W NISKIEJ TEMPERATURZE

Streszczenie. Cięte liście cantedeskii etiopskiej (*Zantedeschia aethiopica* Spr.) i funkii (*Hosta* 'Undulata Erromena') stanowią bardzo wartościowy materiał uprawiany na zieleń ciętą. W pracy sprawdzono wpływ traktowania regulatorami wzrostu zastosowanego przed przechowywaniem w obniżonej temperaturze i sposobu przechowywania liści przez 3 dni w obniżonej temperaturze (5 ± 1°C) (na świetle /na mokro; na świetle /na sucho; w ciemności /na mokro; w ciemności /na sucho), na ich pozbiorczą trwałość oraz na zmiany zawartości chlorofilu w trakcie starzenia. Po przechowywaniu liście umieszczano w fitotronie (20 ±1°C w rytmie dobowym 12 h światła, 12 h ciemności). Wyniki przeprowadzonych doświadczeń potwierdziły wykazany wcześniej negatywny wpływ przechowywania ciętych liści cantedeskii etiopskiej w ciemności na sucho, a liści funkii na sucho (zarówno w ciemności, jak i na świetle) na ich dekoracyjność. Stwierdzono jednak, że negatywne skutki przechowywania ciętych liści obu gatunków w ciemności bądź na sucho można przynajmniej częściowo zniwelować, stosując przed przechowywaniem 24-godzinne kondycjonowanie regulatorami wzrostu: w przypadku liści cantedeskii – kwasem giberelinowym, a w przypadku liści funkii – benzyloadeniną. U cantedeskii zabieg ten całkowicie niweluje negatywne skutki przechowywania w ciemności w perforowanej folii polietylenowej, utrzymując ich trwałość na poziomie 7-krotnie wyższym od liści nietraktowanych. U liści funkii kondycjonowanie benzyloadeniną zwiększa ich trwałość 6-krotnie w stosunku do liści nietraktowanych. Mimo tak dużego wzrostu trwałości, w liściach funkii traktowanych benzyloadeniną utrzymuje się negatywny efekt przechowywania na sucho. Korzystny wpływ regulatorów wzrostu odzwierciedlił się w wyższym poziomie chlorofilu w liściach.

Słowa kluczowe: *Zantedeschia aethiopica*, *Hosta* 'Undulata Erromena', przechowywanie, regulatory wzrostu, starzenie, zieleń cięta

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