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POST-HARVEST LONGEVITY OF LEAVES OF THE IBERIAN CRANESBILLS (*GERANIUM PLATYPETALUM* FISCH. ET MEY.) AFTER THE APPLICATION OF GIBBERELLIC ACID

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Abstract

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In the following study the influence of *Geranium platypetalum* leaves conditioning in gibberellic acid onto their post-harvest longevity and quality was assessed. Leaves were conditioned in water solution of gibberellic acid (GA_3) at the concentration of 25 and 50 ppm for 4 and 24 hours. 4-hour leaves conditioning was conducted in a room at the temperature of 18-20°C, and with 10-hour photoperiod and under fluorescent light of quantum irradiance of 25 $\mu mol \cdot m^{-2} \cdot s^{-1}$. 24-hour leaves conditioning was conducted in a cool room at the temperature of 5°C. Afterwards, the leaves were placed in distilled water. Leaves from the control group were placed in distilled water immediately after harvesting. 24-hour conditioning of *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 and 50 ppm prolonged their post-harvest longevity by 9.4 – 23.2 days. Gibberellic acid had a retarding effect onto chlorophyll decomposition in leaves. 4 and 24-hour conditioning of *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 and 50 ppm has a favourable effect onto protein content at different stages of the advancing ageing process. Conditioning of *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 ppm for 24 hours and in GA_3 at the concentration of 50 ppm increased the saccharides content.

Key words: florists' greens, growth regulators, index of leaves greenness, longevity, protein, saccharides

Introduction

Conditioning is a simple and effective method of post-harvest longevity of both flowers and florist greens. Performing this treatment immediately after harvesting by the producer increases the quality of floral products expressed as their post-harvest longevity.

Conditioning usually takes between 4 and 24 hours in a room at the temperature of 18-20°C or in a cool room.

Due to the differences in ageing processes in leaves as compared to flowers, standard mediums are usually not very effective and they even lower the decorative values. Hence experiments with growth regulators from gibberellin and cytokinin group are carried out in order to improve the post-harvest longevity of florists' greens (Janowska, 2010; Janowska and Jerzy, 2003; Janowska and Schroeter-Zakrzewska, 2008; Janowska and Śmigielska, 2010; Skutnik et al., 2004).

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Perennial leaves can be perceived as an alternative for the species cultivated under covers as the costs of their productions are significantly lower.

In the conducted experiment the influence of *Geranium platypetalum* leaves conditioning in gibberellic acid onto their post-harvest longevity and quality was assessed.

Materials and Methods

Leaves of Iberian cranesbill (*Geranium platypetalum* Fisch. et Mey.) were used in the experiment. They were obtained in the early morning hours from the collection of the department's ornamental plants. The leaves selected for the experiment were fully developed, without any damage or discolouration.

The leaves were conditioned for either 4 or 24 hours in water solutions of gibberellic acid (GA_3) at the concentration of 25 and 50 ppm. Gibrescol 10MG, containing 10% of gibberellic acid, was used in the conditioning process. 4-hour conditioning was conducted in a room at the temperature of 18-20°C, with 10-hour photoperiod and under fluorescent light of quantum irradiance of 25 $\mu mol \cdot m^{-2} \cdot s^{-1}$. 24-hour conditioning was conducted in a cool room at the temperature of 5°C. After the conditioning process the leaves were place in distilled water. The leaves from the control treatment were placed in distilled water immediately after harvesting.

The experiment consisted of 6 treatments in both years of the study (for vase life and index of leaves greenness). One treatment (conditioning time × GA_3 concentration) comprised 15 leaves, 5 in 3 different replications. For the protein and the saccharides contents the experiment consisted of 6 variants (date) in GA_3 concentration and in conditioning time.

Leaves post-harvest longevity was determined in a room at the temperature of 18-20ºC, with 10-hour photoperiod and under fluorescent light of quantum irradiance of 25 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Air relative humidity was kept at the level of 70%.

Post-harvest longevity was expressed in days. The moment when 30% of the leaf surface was yellowed or withered meant the loss of ornamental value. The index of leaves greenness (SPAD), correlated with chlorophyll content (Gregorczyk and Raczyńska, 1997; Gregorczyk et al., 1998) (with the use of N-Tester apparatus) and protein and saccharides contents were determined.

The determination of protein content in the leaves was made with the help of Bradford's (1976) method. 2ml of a solution of Coomassie Brilliant Blue G-250 (CBB) in 85% orthophosphoric acid was added to 100 µl of a diluted extract, with the extraction in a phosphorate-potassium buffer (pH 7.0). After 10 minutes the absorbance was measured at a wavelength of 595 nm. Protein content was determined from a curve plotted for albumin.

Total saccharides were determined using the antron reagent (Brörnesjo, 1955). Under the effect of sulphuric acid, saccharides transform into derivatives of furfural, which, together with antron, yield blue-green products. The intensity of the colour is proportional to their content. Weighed portions (0.5 g) were crushed in a mortar with 5 cm^3 of distilled water and the homogenate was centrifuged at 10,000 g for 20 minutes. 1 cm^3 of the supernatant was added to 2 cm^3 of a cooled antron reagent (0.02% in concentrated H_2SO_4), and then the contents of the test tubes were heated, while slowly mixed, in a water bath at 90°C for 14 minutes. After the tubes were cooled, the absorbance of the solutions was measured in a spectrophotometer at a wavelength of 620 nm. The content of saccharides was read from a standard curve prepared for glucose. The final results, which were means of four replications, were expressed in mg of glucose per g of fresh weight.

Experimental data were subjected to a one-factor (for the protein and saccharides contents) or three-factor (for vase life and index of leaves greenness) analysis of variance (Anova) and significant differences between means were determined by Tukey multiple range test. Data significantly different from respective control are expressed on the figures as asterisks * $P< 0.05$, ** $P< 0.01$.

Results

Post-harvest longevity of *Geranium platypetalum* leaves was significantly dependent of both, the used solution of gibberellic acid and the length of the conditioning process (Table 1). Regardless of the solution of gibberellic acid the highest longevity was exhibited by the leaves conditioned for 24 hours. Simultaneously, regardless of the length of the conditioning process the highest longevity was exhibited by the leaves that were conditioned in a solution of gibberellic acid at the concentration of 50 ppm. Comparing the interactions it must be assumed that the leaves exhibited the highest longevity after they were conditioned for 24 hours in the solution of GA_3 at the concentration of 50 ppm. When compared with the leaves from the control group, their longevity increased by 23.2 days. The leaves that were conditioned for 24 hours in the solution of gibberellic acid at the concentration of 25 ppm also exhibited significantly higher longevity. It was by 9.4 days longer than the longevity exhibited by the leaves from the control group.

The index of leaves greenness of *Geranium platypetalum* was significantly and solely dependent on the length of the conditioning process (Table 1). It was proved that, regardless of the concentration of gibberellic acid, the higher index of leaves greenness was exhibited by the leaves conditioned in GA_3 for 4 and 24 hours, with no significant differences as far

Table 1
Influence of conditioning in gibberellic acid onto post-harvest longevity and index of leaves greenness of *Geranium platypetalum* leaves

Year	Conditioning (h)	GA_3 concentration (ppm) 25	GA_3 concentration (ppm) 50	Mean for conditioning
		Vase life (days)		
2011	0	11.0	11.0	11.8
2012		12.5	12.5	
2011	4	12.2	12.7	12.3
2012		13.2	11.0	
2011	24	19.5 *	32.0 **	27.3 *
2012		21.9 *	35.7 **	
Mean for conditioning´GA_3 concentration	0	11.8	11.8	
	4	12.7	11.9	
	24	20.7 *	33.9 **	
Mean for GA_3 concentration		15.1	19.2 *	
Year	Conditioning (h)	GA_3 concentration (ppm) 25	GA_3 concentration (ppm) 50	Mean for conditioning
		Index of leaves greenness (SPAD)		
2011	0	29.8	29.8	30.5
2012		31.1	31.1	
2011	4	37.6 *	38.0 *	38.6 *
2012		38.9 *	40.0 *	
2011	24	37.0 *	37.6 *	38.1 *
2012		36.9 *	41.0 *	
Mean for conditioning´GA_3 concentration	0	30.5	30.5	
	4	38.2 *	39.0 *	
	24	37.0 *	39.3 *	
Mean for GA_3 concentration		35.2	36.2	

*P<0.05, **P<0.01

as the length of this process is concerned. While comparing the interaction, the lowest value of the index of leaves greenness was exhibited by the leaves from the control treatment. The leaves conditioned in gibberellic acid at both mentioned above concentrations exhibited higher values of the index of leaves greenness.

The changes in the protein content in *Geranium platypetalum* leaves during the advancing ageing process were observed (Table 2). In the control combination protein content changes were small with the exception of day two of the experiment when the protein level increased significantly. In the combination in which the leaves were conditioned in GA_3 at the concentration of 25 ppm for 4 hours, initially the protein content remained almost unchanged but then it increased significantly. In the case of the leaves conditioned in GA_3 at the concentration of 25 ppm for 24 hours protein content continued to grow gradually. As far as the leaves conditioned in gibberellic acid at the concentration of 50 ppm for either 4 or 24 hours are concerned, the growth in the protein content was observed at the beginning of the experiment.

During the advancing ageing process of *Geranium platypetalum* leaves the changes in saccharides content were observed (Table 2). Statistically insignificant differences were observed in three treatments: control, the treatment in which the leaves were conditioned in gibberellic acid at the concentration of 25 ppm for 4 hours, and the one in which gibberellic acid at the concentration of 50 ppm was used for 24-hour conditioning.

The significant increase in the content of saccharides in the leaves of the species in question during the ageing process was observed in the leaves conditioned in gibberellic acid at the concentration of 25 ppm for 24 hours. Similarly, the significant increase in the content of saccharides at the beginning of the age-

Table 2
Influence of conditioning in gibberellic acid onto total protein and saccharides content of *Geraniujm platypetalum* leaves

Conditioning (h)	GA_3 concentration	Date					
	(ppm)	13.06	15.06	18.06	20.06	22.06	25.06
mg protein·g^{-1} FW							
0		1132	1431**	1092	1018	1129	1129
4	25	948	963	960	1212**	1358**	811
24		824	1008**	1060**	1176**	1241**	1180**
0		1132	1431**	1092	1018	1129	1129
4	50	1098	1335**	1182	1124	1217*	1161
24		945	1088	1081	1063	1070	768
mg glucose·g^{-1} FW							
0		44.91	45.42	50.70	40.50	41.32	33.52
4	25	40.9	45.72	45.24	45.10	47.08	25.70
25		30.70	42.36**	42.37**	48.55**	44.64**	26.09
0		44.91	45.42	50.70	40.50	41.32	33.52
4	50	36.5	44.16**	50.30**	33.19	41.58	26.83
24		40.40	37.08	38.50	43.50	43.90	29.01

*P<0.05, **P<0.01

ing process was observed in the leaves conditioned in gibberellic acid at the concentration of 50 ppm for 4 hours.

Discussion

In the conducted experiment *Geranium platypetalum* leaves from the control treatment kept their longevity for 12.5 days. Conditioning leaves for 24 hours in gibberellic acid at the concentration of 25 and 50 ppm increased their post-harvest longevity for 9.4 – 23.2 days. Moreover, conditioning leaves in gibberellic acid had a retarding effect onto chlorophyll decomposition. The effectiveness of gibberellic acid used in increasing post-harvest longevity of florists' greens is reported by Janowska and Schroeter-Zakrzewska (2008). The authors obtained more durable after harvesting leaves of *Arum italicum* due to conditioning them in gibberellic acid. Moreover, gibberellic acid had a retarding effect onto chlorophyll decomposition in ageing leaves of this species. According to the study by Janowska and Stanecka (2011) the increased post-harvest longevity of *Zantedeschia* 'Sunglow' leaves was also obtained by means of conditioning them in gibberellic acid at the concentration of 400 ppm Moreover, gibberellic acid at the concentration of 300-400 ppm had a retarding effect onto chlorophyll decomposition in leaves. Similarly, in the case of 'Black Eyed Beauty' cultivar gibberellic acid proved to be effective, as, when applied at the concentration of 50 and 100 ppm, it increased the post-harvest longevity of the cut leaves and had a favourable effect onto chlorophyll content in leaves. In the study by Janowska and Jerzy (2003) gibberellic acid had a favourable effect onto the longevity of *Zantedeschia* 'Florex Gold' and 'Black Magic' cut leaves. The leaves of these cultivars kept their ornamental value for the longest period of time when they were conditioned in the solution of gibberellic acid at the concentration of 300 ppm. Comparable longevity was exhibited by 'Florex Gold' cultivar leaves placed in water and, prior to that, conditioned in gibberellic acid at the concentration of 200 ppm. Increasing longevity was combined with chlorophyll decomposition retardation, which is why the leaves kept their green colour for a longer period of time. Similarly, in the case of *Zantedeschia aethiopica* leaves, gibberellic acid increased their post-harvest longevity six fold (Skutnik et al. 2001). In the study by Skutnik et al. (2004) gibberellic acid retarded chlorophyll decomposition in *Zantedeschia aethiopica* and *Zantedeschia elliottiana* leaves, simultaneously, increasing their post-harvest longevity. In the study by Janowska and Schroeter-Zakrzewska (2010) *Limonium latifolium* leaves placed into water kept their ornamental values for 6 days. The application of gibberellic acid at the concentration of 25 and 50 ppm to the leaves conditioning process increased their post-harvest longevity in a significant way. What is more, conditioning leaves in gibberellic acid had a retarding effect onto chlorophyll decomposition. Gibberellin applied in *Alstroemeria* (Dai and Paull, 1991; Hicklenton, 1991) and *Lili-*

um (Han, 1995; Rabiza-Świder et al., 2012) retards effectively chlorophyll decomposition in leaves. It is also benzyladenine, apart from gibberellins, that is used in the process of increasing post-harvest longevity of florists' greens. Its effectiveness was proved in numerous species (Janowska and Schroeter-Zakrzewska, 2008; Skutnik and Rabiza-Świder, 2005; Skutnik et al. 2006). Sometimes mixtures containing cytokinins and gibberellins are used to increase post-harvest longevity of florists' greens. In the study by Janowska et al. (2012) postharvest longevity of *Zantedeschia albomaculata* 'Albomaculata' leaves was significantly dependent on both the concentration of *meta*-methoxytopolin and gibberellic acid, and on the conditioning method. The leaves, in the case of which a few-second dipping of leaf blades was applied, regardless of the concentration of growth regulators, kept their ornamental value for a longer period of time. After the application of growth regulators at the concentration of 25+25 ppm to a few-second leaf blades dipping the postharvest longevity increased by 12.3 days, and in the case of the concentration of 50+50 ppm it increased by 23.9 days. The authors also proved a favourable effect of the concentration of the mixture of MemT+GA_3 onto the index of leaves greenness. The durability of the leaves of the examined species increased also after the application of the mixture of *meta*-methoksytopolin riboside and gibberellic acid to the process of a few-second leaf blades dipping.

4 and 24-hour conditioning of *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 and 50 ppm has a favourable effect onto the protein content at different stages of the advancing ageing process.

The consequence of the advancing process of leaf senescence is proteolysis, i.e. the degradation of proteins. There is little information in the available literature on the inhibition of this process after the use of growth regulators. In the research by Rabiza-Świder et al. (2004), leaves of *Zantedeschia aethiopica* and *Zantedeschia elliottiana* were subjected to 24-hour conditioning in solutions of benzyladenine and gibberellic acid. In both species only gibberellic acid effectively retarded the degradation of soluble proteins. The standard medium employed to extend the longevity of cut flowers accelerated proteolysis in leaves of *Z. aethiopica*, but did not show the same unfavourable effect on those of *Z. elliottiana*. A decline in the content of soluble proteins was accompanied by the accumulation of free amino acids. The favourable effect of gibberellic acid onto the protein content in the leaves of calla with colourful inflorescence spathes was also observed by Janowska and Stanecka (2011) and Janowska et al. (2012). Similarly, in a study by Rabiza-Świder and Skutnik (2008), the conditioning of leaves of *Hosta* 'Crispula' and 'Undulata Mediovariegata' in gibberellic acid and benzyladenine retarded the degradation of soluble proteins, especially readily visible after the use of benzyladenine. In turn, placing *Hosta* leaves in the standard medium used for cut flowers accelerated proteolysis.

Conditioning *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 ppm for 24 hours and in GA_3 at the concentration of 50 ppm influences the increase in the content of saccharides.

Sugars that are created in the photosynthesis process are the major structural and storage material in plant organisms. Intensive photosynthesis results in storing greater amounts of carbohydrates. In the available literature there is very little information on the changes in sugar content in ornamental plants that are due to the application of growth regulators. Kozłowska et al. (2007) report about the changes of sugar content in the leaves of *Zantedeschia elliottiana* after the application of gibberellic acid to rhizome soaking depending on their development phase. The authors report that during the initial phase of vegetative development the leaf blades of the plants treated with gibberellic acid were characterised by the higher content of hydrocarbons, especially fructose and glucose when compared with the plants from the control treatment. This content increased together with the leaves development but decreased when the plants entered the generative stage. At that time the total content of hydrocarbons in the leaves of the control plants was twice as high. The changes in the sugar content in cut leaves of *Zantedeschia aethiopica* and *Z. elliottiana* were examined by Skutnik et al. (2004). The content of reducing sugars during the advancing ageing process increased at the beginning and then fell to 60-80% of the initial level. Conditioning the leaves in the solution of benzyladenine did not have a retarding effect onto this process. Gibberellic acid proved to be more effective as it delayed sugar degradation in the leaves of *Zantedeschia aethiopica*, and in *Z. elliottiana* it resulted in sugar content increase. Downs et al. (1997) report about a favourable effect of benzyladenine onto the sugar content in broccoli inflorescences as it retarded their decomposition that follows harvesting. Smoleń and Sady (2009), on the other hand, proved a favourable effect of benzyladenine onto the sugar content in the fleshy part of the radish taproot.

Conclusion

In conclusion, 24-hour conditioning of *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 and ppm increased their post-harvest longevity by 9.4 – 23.2 days. Gibberellic acid had a retarding effect onto chlorophyll decomposition in leaves. 4 and 24-hour conditioning of *Geranium platypetalum* leaves in gibberellic acid at the

concentration of 25 and 50 ppm has a favourable effect onto the protein content at different stages of the advancing ageing process. Conditioning *Geranium platypetalum* leaves in gibberellic acid at the concentration of 25 ppm for 24 hours and in GA_3 at the concentration of 50 ppm increases the saccharides content.

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