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Using of preservative solutions to improve postharvest life of *Rosa Hybrid* cv. Black magic

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Ornamental products such as cut flowers have a limited display life so the effect of some effective chemical holding treatments on the vase life of rose (*Rosa hybrida* L.) cv. Black magic cut flowers was studied. In order to find the best treatment in extending cut rose flowers vase life, cut roses were kept in preservative solution including 2.5% sucrose supplemented with 8-hydroxyquinoline citrate, copper sulfate, silver nitrate, aluminum sulfate, ethanol and water (control). All treatment flowers were stored in the climate controlled room at 20±2°C with 90% RH and 12h photo period using fluorescent lamps (light intensity of 15µmol $m^{-2}s^{-1}$) at the top of corolla .The result showed that ethanol and aluminum sulfate treatments had the most important role in the extending longevity as well as water uptake in these treatments. Also, there was no significant difference between treatments for leaf and petal dry weight, but the partially high level of water content in aluminum sulfate and ethanol treatments can be effective in increasing vase life. The amount of anthocyanin leakage was high in control. Controls vase life ended at day 9, while the vase life of flowers was kept in aluminum sulfate and ethanol were 18 day.

Key words: rose (*Rosa Hybrid* L.), postharvest quality, holding solutions, vase life

Introduction

Roses are one of the most important cut flowers in the world (Şirin, 2011) and extremely perishable (Figueroa *et al*., 2005) as well as other cut flowers such as Eustoma grandiflorum (Hojjati *et al.*, 2007; Farokhzad et al., 2005), Gerbera jamesonii (Nair *et al*., 2003). Short postharvest vase life is one of the most important problems in cut flowers (Zamani *et al*., 2011). So consider to maintaining postharvest quality of cut flowers is critical for preventing of

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flower post harvest losses. Senescence which is the main factor affecting on flower quality can be induced by several per and post- harvest factors e.g., water stress (Sankat and Mujaffar, 1994), amount of carbohydrates (Coorts, 1973; Ketsa, 1989), microorganisms (Van Doorn and Witte, 1991), ethylene effects (Wan & Miller, 2003) as documented in carnation and roses (Mayak and Halevy, 1980; Halevy and Mayak, 1981; Quesada and Valpuesta, 2000) and Lisianthus (Farokhzad *et al*., 2005; Hojjat *et al*., 2007) and cultivar differences, season, development stage at harvest and cultivated conditions (Doel and Wilkins, 1999). The senescence of cut flowers is closely related to the considerable reduction of the energy needed for synthesis reactions (Figueroa *et al*., 2005), it seems that supply of exogenous carbohydrate should be enough to delay the senescence (Kaltaler & Steponkus, 1976) and provide a respiratory substrate (Cho *et al*., 2001). However, carbohydrate especially sucrose cause to growth and increase in bacterial amounts, existing in preservative solution, which tends to embolism of cut flower xylem vessels, consequently (Stephens *et al*., 2001). On the other hard ethylene is recognized for petal senescence (Halevy & Mayak, 1981) which is accompanied by the irreversible in rolling, wilting and abscission of the petals (Kim *et al*., 2005). Addition of chemical preservatives to the holding solution is considered a common practice for the storage of floral stems. Thawiang *et al* (2007) suggested that the most effective treatment in extending vase life of Anthorium is ethanol up to 2%. As described above all preservative solution must essentially contain two components including sugar and germicides (Nair and Sharna, 2003). These treatments allow to control the ethylene synthesis (Arboleda, 1993), photogene development (Nair and Sharna, 2003) and flower opening (Hojjati *et al*., 2007). A large member of bactericides such as aluminum sulphate, silver nitrate, silver thiosulphate and sodium thiosulphate are available for using in the holding solutions (Figueroa *et al*., 2005). It is demonstrated that silver thiosulphate is a competitor inhibitor of ethylene (Nowake and padnicki, 1990; Arboleda, 1993). Silver thiosulphate can delay petal senescence in the most of cut flowers. Figueroa *et al* (2005) concluded that pre treatment with silver nitrate on rose cvs. Raphaells and Bettira tend to decrease in the content of soluble and reducing sugars in the petals but sucrose added to the preservative solution compensate their decrease in petals. Using of 8-hydroxy quinolin salts are very common chemicals used in roses as a floral preservative (van Doorn, 1997).

Different preservatives were used for improving postharvest physiochemical parameters of Eustoma grandiflorum such as BA (hassanpour asil and karimi), ethanol (Farokhzad *et al*., 2005), Aluminum sulphate (Liao et al., 2001) and 8-hydroxy quinolin citrate, cobalt chloride, copper sulphate and citric acid (Hojjati *et al*., 2007). It has been reported that in gerbera gamesonii application of 20 ppm $AgNo_3$ + 4% sucrose was more effective treatment in extending vase life (5). Ethanol is one of the most effective chemicals in preserving quality of carnation by inhibiting ethylene biosynthesis (Wu *et al*., 1992; Pun *et al*., 1999) and its action (Wu *et al*., 1992). The purpose of present study was to evaluate the physiological effects of different compounds used to formulate flowery improving post harvest quality of *Rosa hybrida* L. cv. Black magic.

Materials and methods

Plant material, flower stems of roses (*Rosa hybrida* L.) cv. Black magic with partially short vase life (Hajizadeh *et al*., 2011) were purchased at harvest time from a commercial grower in Tehran-Iran. Flower stems were trimmed to 45cm and were placed (5 cut flowers) in a 500 *ml* flask with 250 *ml* of solution. Experiments were arranged in completely randomized design with three replications. Cut flowers placed in vase solution containing as follows:- T_1=8-hydroxyquinoline citrate (300 ppm), T_2=copper sulphate (100 ppm), T_3=silver nitrate (30 ppm), T_4=aluminum sulphate (100 ppm), T_5= Ethanol 2% and T_6= control (Distilled water). Sucrose is one of the most effective compound in preservative solutions (Han, 2003; Ichimora and korenaga, 1998). It was used the moderate percentage of sucrose in all solutions but not alone because of some microbial problems. Hence 2.5% sucrose was added in the all treatments except for controls. All treatments were placed in chambers at room temperature 20±2 ˚C. The relative humidity was about 70% while 12h photoperiod was maintained using fluorescence lamps whit a light intensity of 15-20 $\mu mol\ m^2 s^{-1}$ at the top of corolla. Another flask with 250 ml Distilled water and without flower set up for calculating the rate of evaporation. Data were statistically analyzed using SAS software. The vase life of cut roses was ended when the petals lost their turgidity and bent neck was occurred.

Physiochemical parameters

Rate of bud opening (flower diameter), water uptake, chlorophyll, anthocyanin, dry weight of leaf and petals, water content of leaves and petals and vase life were recorded each 3 days in all treatments. The fresh weight of the cut flowers also measured every 3 days during sampling times for calculating changes in fresh weight during vase life.

Chlorophyll content

Total chlorophyll content was measured by chlorophyll meter (SPAD-502, Minolta Co. Japan). Average of 3 measurements from different spots of a single leaves was considered.

Water uptake

The volume of water uptake was calculated by subtracting the volume of water evaporated from a control bottle without cut flowers from the amount of water decreased in bottles containing flowers.

Changes in fresh weight and dry weight of leaves and petals

Cut flowers were weighted and the change in fresh weight was calculated as a percentage on basis of initial fresh weight. Petals and leaves from each treatment were dried in oven at 80° C till constant weight to determine dry mass. Change in dry weight was calculated at a percentage on basis of initial dry mass.

Anthocyanin leakage

To determine the effect of different chemical solutions on anthocyanin leakage, petal slices was extracted with methanol containing 1% HCL overnight at 4° C. The absorbance of the extracts was measured at 530-700 *nm* with a spectrophotometer (Murr *et al*, 2008).

Statistical analysis

This experiment was conducted in a completely randomized design with tree replications. 5 stems of rose cut flowers were used for each replication. Results were analyzed by using SAS software. Mean comparisons to identify significant difference between treatments were performed using Duncan's multiple range tests at the 5% level.

Results and discussions

Vase life - Results showed a significant difference between all different chemical holdings and control from vase life point of view and as documented in Figure 1. All treatments except for T2 (Copper sulphate) were effective in extending vase life of cut rose flowers in comparison to controls. The vase life of cut rose flowers in the control treatment were only 9 days whereas flowers treated with ethanol and specially Aluminum sulphate preservatives extended vase life of flowers up to 18 days. Our results for the potential of ethanol in extending flower longevity are agreed with Karimian and Tehranifar (2011). Ethanol by decreasing in ethylene production and or sensitivity to ethylene and also as an antimicrobial compound can prolong some cut flowers vase life as reported by Heins and Blakely (1980) and Wu *et al* (1992).

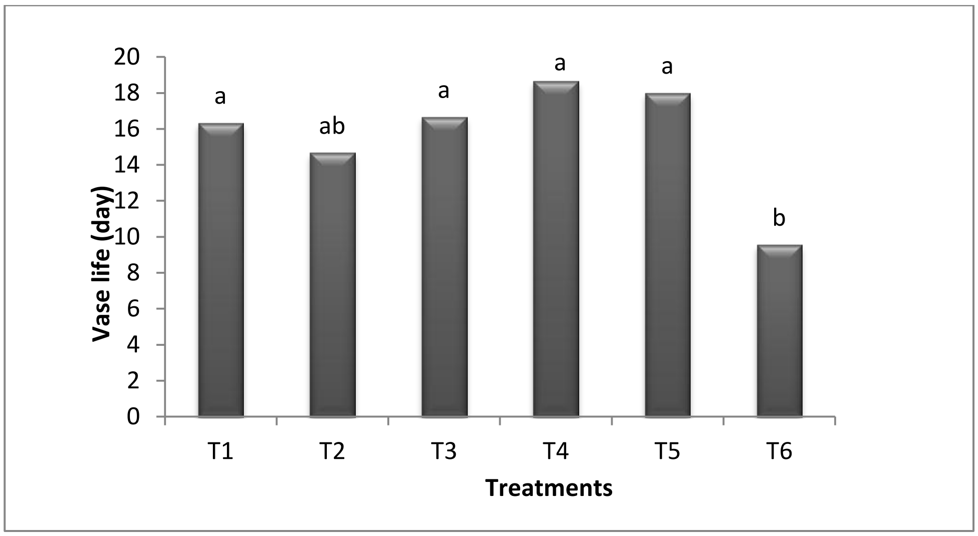

Fig. 1. The effect of different preservative treatments on the vase life of cut rose flowers cv. Black magic. T_1= 8-Hydroxy quinolone citrate, T_2= Copper sulphate, T_3= Silver nitrate, T_4= Aluminum sulphate, T_5= Ethanol and C= Control.

Water uptake - The results showed that there is a significant difference between treatments and control in levels of water uptake as the level of water uptake has a decreasing trend in all treatments (Figure 2). Water uptake of cut rose in control reduced gradually with the progress of vase life until day 9 whereas the flowers placed in solution with ethanol and aluminum sulphate had high water uptake that were the best factor in extending vase life of cut rose flowers. Our results about the increasing in water uptake in solutions including ethanol are agreed with Farokhzad *et al*, 2005. It is clear that the presence of microorganisms in vase solutions can cause physical plugging of cut stems, release toxic metabolites and result in programmed cell death (PCD) (Alvarez, 2000). Ethanol is a disinfectant which can improve water conductance by preventing bacterial growth and stem embolism. Higher water uptake in solutions including aluminum sulphate (van Doorn, 1998) and ethanol allowed more increase in fresh wieght and mainly due to better corolla development.

Water balance is a major factor determines quality and longevity of cut flowers. It is influenced by water uptake and transpiration, being balance between these two processes (da Silva, 2003). Low water uptake is often due to occlusions located mainly in the basal stem end (He *et al*., 2006) and microbes are common cans of stem end blockage (van doorn, 1997). Many agents have been used in cut flowers vase solution to extend vase life by improving water uptake such as silver nitrate (Fujino *et al*., 1983), aluminum sulphate (Ichimura and shimizu-yumota, 2007) and 8-hydroxyquinolin (Ichimura *et al*., 1999).

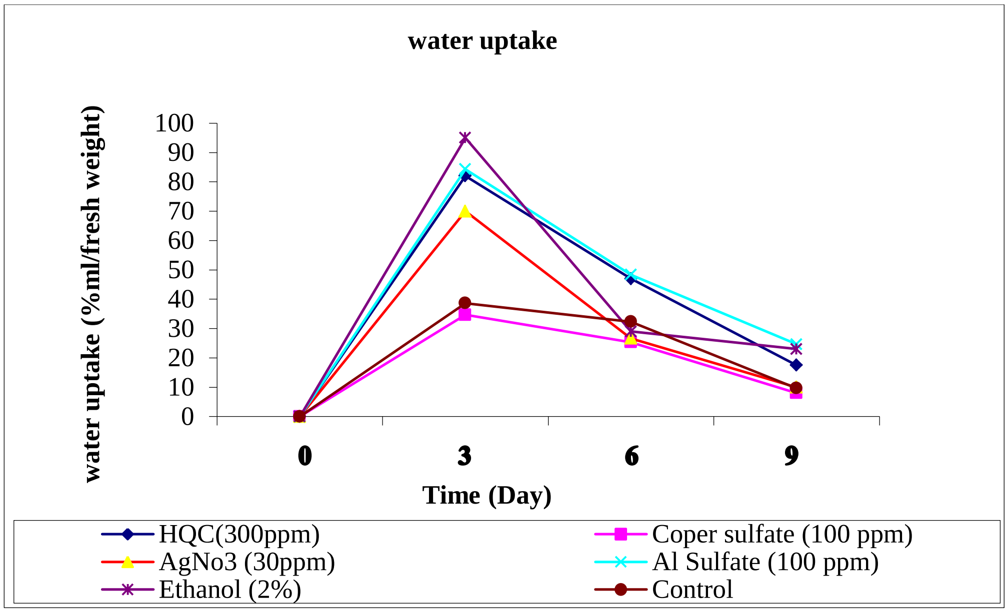

Fig. 2. The effect of different preservative treatments on water uptake of cut rose flowers cv. Black magic. T_1= 8-Hydroxy quinolone citrate, T_2= Copper sulphate, T_3= Silver nitrate, T_4= Aluminum sulphate, T_5= Ethanol and C= Control.

Dry weight and water content- There was no significant difference between treatments in leaf dry weight and leaf water content (Table 1). While there was a significant difference between petals dry weight placed in aluminum sulphate and ethanol with control as control flowers have more dry weight than other treatment. On the other hand no significant difference showed between treatments and control in petal water content, however petal water content in control was less than others, specially aluminum sulphate and ethanol treatments. It seems that control flower petals with less water content lose their internal water faster than other treatments which lead to their wilting and short vase life. It is suggested that more flower water content save flower display life for more time.

Relative fresh weight- Results showed a significant difference between flowers treated with Ethanol and controls as ethanol treatment saved more fresh weight in flowers than control (Table 1). Relative fresh weight of flowers had a decreasing trend during vase life and the most decrease in relative fresh weight observed in controls at the end of vase life (9 days) (Table 1). As a consequence with decreasing flower fresh weight losses, vase life will increase as shown in flowers treated with ethanol with 18 days vase life. Extending vase life of cut flower depends absolutely on adequate supply of water and any problem in water uptake can result in rapid wilting of stem, petal and leaves and decrease in fresh weight which accounts as the onset of senescence (Adachi et al, 2000; Ichimura and Goto, 2002).

Bud opening- Bud opening was determined by following changes in flower diameter. As shown in table 1 there was a significant difference between flowers placed in 8-HQC and Aluminum sulphate solutions with controls as un treated cut flowers had a less diameter comparison to HQC and aluminum sulphate. Flowers placed in Aluminum sulphate solution had a high flower diameter. It seems that high water uptake, supply of sucrose and lack of contamination in this treatment allow to flower opening until the end of vase life as terminated at day 18 (Table1).

Chlorophyll- Results showed a significant difference between two preservative solutions including aluminum sulphate and ethanol with control and copper sulphate solution as the minimum total chlorophyll content was noted in control and copper sulphate solution. The flower kept in aluminum sulphate and ethanol lead to a considerable delay in degradation of chlorophyll compare to control and copper sulphate as shown in Table 1. In this case, aluminium sulphate and ethanol solutions were better than other treatments and it seems that the rate of transpiration and deterioration is very slow in these two treatments.

Anthocyanin leakage- The results indicated that flowers placed in preservative solutions including 8-HQC and silver nitrate cause to significant decrease in anthocyanin leakage compared to control (Table 1). It seems that membrane stability with increased concentration of anthocyanin leakage in controls compared to treatments 8-HQC and silver nitrate was higher in these treatments.

Table 1. The effect of different preservative solutions on quality parameters of rose cut flower cv. Black magic

Treatments	Quality parameters							
	Chlorophyll content	Anthocyanin Leakage (mgL^{-1}) (absorption at 525*nm*)	Bud opening rate	Leaf dry weight Mg/g FW	Leaf water content Mg/g FW	Petal dry weight Mg/g FW	Petal water content Mg/g FW	Relative fresh weight
8-HQC (300 ppm) +2.5% sucrose	57.108 ^{AB}	146.25 ^{A}	36.545 ^{A}	192.50 ^{A}	2.239 A	149.17 AB	2.117^{A}	97.033 ^{AB}
Copper sulphate (100 ppm) + 2.5% sucrose	49.417 ^{C}	166.92 ^{AB}	33.114 ^{AB}	207.50 ^{A}	1.136 A	167.5^{A} B	1.736 A	94.880 ^{AB}
Silver nitrate (30 ppm) +2.5% sucrose	54.00 ^{BC}	143.42 ^{A}	31.408 ^{AB}	182.50 ^{A}	1.966 A	155.00 AB	1.566 A	95.905 ^{AB}
Aluminum sulphate (100 ppm) +2.5% sucrose	57.342 ^{A}	160.25 ^{AB}	43.446 ^{A}	140.0 ^{A}	1.235 A	120.0^{B}	2.894 A	96.476 ^{AB}
Ethanol (2 %) +2.5% sucrose	58.338 ^{A}	169.25 ^{AB}	28.926 ^{AB}	142.50 ^{A}	0.981 A	118.33 B	3.406 A	98.874 ^{A}
Control	52.229 ^{C}	180.29 ^{B}	26.147 ^{B}	168.57 ^{A}	1.202 A	194.29 A	1.438 A	89.431 ^{B}

Results showed that the application of different chemical preservatives were effective in extending vase life of cut rose flowers in comparison with controls as documented in Table 1. In this case, results showed that the application of different chemical preservatives delayed the chlorophyll and anthocyanin degradation in comparison to untreated control. Leaf yellowing is characterized by breakdown of chlorophylls, proteins and nucleic acid in the detached leaves (Kofranek & Halvey, 1981). The best treatments in this regards were ethanol and aluminum sulfate (Table 1). It seems that all chemicals plus sucrose had the important role in preventing of yellowing & finally senescence which was observed rapidly in control after 9 days (Table 1). Similar results were obtained by using of plant growth regulators on leaf senescence decay and quality of cut flowers were improved (Emongor *et al*, 2000; Mutui *et al*, 2001).

Color lacking and discoloration are important factors in determining display life of cut flowers and almost determine end of vase life. Two major types of pigments involved in flowers color including carotenoids and anthocyanin (Amarjitt, 2000). It seems that low levels of carbohydrate, high respiration rate and ethylene production can cause to petal color lacking. As it is not absolutely clear that *Rosa hybrida* L. cv. Black magic is sensitive to ethylene or not, so our suggestion mainly is relies on the role of different chemicals in decreasing of respiration rate, Aluminum sulphate, silver nitrate (Nowak and Rudnicki, 1990) and ethanol are among the most common bactericides and have the possibility to prevent of growth and development of bacteria and ethylene production consequently. On the other hand silver compounds compete with ethylene for the same site of action in ethylene transduction pathway (Arboled, 1993), so anthocyanin was preserved in comparison to control.

Conclusion

It can be concluded that using of ethanol and aluminum sulfate along with sucrose has a great potential to extend the vase life of rose cut flowers up to 18 days which done by decreasing fresh weight losses trend and preserving leaves chlorophyll, increasing water uptake and retarding anthocyanin degradation. However, more investigations is needed to find the best solution as floral preservative to preserve postharvest quality parameters of cut *Rosa hybrida* L. cv. Black magic. However, sucrose alone leads to microbial growth, hence the combination of sucrose and biocides might have prolonged the vase life of cut flowers. Among all treatments preservative solutions including ethanol and aluminum sulphate were the best floral solutions as compared to control with regard to vase life. Totally 2% ethanol and 100 ppm aluminum sulphate had better performance in preventing of wilting and senescing of cut rose flowers.

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