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INFLUENCE OF CITRIC ACID AND HYDROGEN PEROXIDE ON POSTHARVEST QUALITY OF TUBEROSE (POLIANTHES TUBEROSA L. 'PEARL') CUT FLOWERS

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ABSTRACT

Quality of cut flowers is an important issue at postharvest as well as an important factor contributing to marketing of and profitability from the tuberose. In this study, the effects of citric acid (CA) and hydrogen peroxide (H₂O₂) added to the vase water on postharvest quality of tuberose cut flowers were investigated. CA was applied in concentrations of 50, 100, 200, 400 mg·dm⁻³ and H₂O₂ in concentrations of 10, 20, 40 and 80 mg·dm⁻³ and distilled water as control treatment. Results showed that both compounds had significant positive effects on solution uptake, wilting and abscission of florets, relative water content, chlorophyll content, and vase life duration. The effects of 100 and 200 mg·dm⁻³ of CA and 20 and 40 mg·dm⁻³ of H₂O₂ proved to be more effective than other treatments. Both compounds increased the vase life of tuberose cut flowers and CA at concentrations 100 and 200 mg dm⁻³ and H₂O₂ at concentrations 20 and 40 mg dm⁻³ doubled this time up to 14-17 days.

Key words: postharvest treatment, postharvest quality, tuberose

INTRODUCTION

Tuberose (Polianthes tuberosa L.) is a perennial bulbous plant of Asparagaceae. Tuberose is a popular and commercially valuable cut flower produced worldwide (De Hertogh & Le Nard 1993; Cuc & Pilon 2007; Cuc et al. 2010; Taha & Eid 2011; Bahadoran et al. 2012). Extending the vase life after harvest is a significant issue that has a major impact on efficient trading of cut flowers and can increase the economic benefits (Weaver et al. 1998). In the cut flowers of tuberose, wilting and burning of florets and bending of the tips of flower spikes are the major problems that reduce the vase life of cut flowers (Jowkar & Salehi 2005). Various factors such as ethylene sensitivity, bacterial contamination, vascular blockage, and oxidative stress cause petal burning or browning, and floret wilting or abscission in cut flowers (Rubinstein 2000; Sankhla et al. 2003; Rattanawisalanon et al. 2003; Rogers

Moreover, various agents such as microbial activities, air obstruction and physiological response to cutting wound, gum aggregation in xylem, latex leakage, and other processes in the wound lead to vascular blockage and prevent water uptake, which reduces the freshness of cut flowers (Rattanawisalanon et al. 2003; van Ieperen et al. 2002; van Meeteren et al. 2005; van Meeteren et al. 2006; Imsabai et al. 2013). Additionally, recent studies on some cut flowers demonstrated that, with decrease in the freshness of cuts, some bacteria aggregate in the vase solution and lead to senescence of cut flowers (Macnish et al. 2008). By using some acids as antibacterial compounds, the frequency of bacteria in the vase solution can be decreased and the vase life of various cut flowers significantly increased, which is supposedly due to the reduction in vascular blockage (Alaey et al. 2011; Shimizu-Yumoto & Ichimura 2010; Mansouri 2012).

2006; Zhang et al. 2011; Shahri & Tahir 2011).

Van Doorn (2010) showed that decrease in the pH of preservation solutions reduced the rate of bacterial growth and increased the water conduction in xylem of cut flowers. Furthermore, a low concentrations of hydrogen peroxide (H₂O₂) was used as a biocontrol agent to counter several abiotic and oxidative stresses during the postharvest time (Macarisin et al. 2010; Peng et al. 2008). H₂O₂ is an essential molecule, which is involved in a plant's growth and development (Neill et al. 2002). An addition of the H₂O₂ into the preservation solution of lily cut flower led to maintaining the cut flower quality and increasing the vase life (Liao et al. 2012).

The aim of this study was to evaluate the effects of citric acid (CA) and H_2O_2 on postharvest characteristics of tuberose cut flowers. To the best of our knowledge, this is the first report on using exogenous H_2O_2 and CA on tuberose cut flowers in order to extend the vase life and maintain the postharvest quality.

MATERIAL AND METHODS

Experimental material

Tuberose plants (*P. tuberosa* cv. 'Pearl') were cultivated in a greenhouse with air temperature set at 25-30 °C during the daytime and 20-25 °C during the nighttime and a relative humidity of 60%. Flower shoots were harvested in the phase of 1-2 opened 1-2 florets. After cutting the flowers from mother plants, they were transferred to the research laboratory in the Agricultural Faculty of Zabol University. The laboratory temperature was controlled at around 18-22 °C and 55-60% relative humidity, and fluorescent light was set for 12-hour lighting. The experiments were carried out with no stem recutting and no changing of the vase solutions during three weeks.

Postharvest treatments

Flower shoots were placed separately in 200ml bottles containing the CA (50, 100, 200, and 400 mg \cdot dm⁻³), H₂O₂ (10, 20, 40, and 80 mg \cdot dm⁻³), or distilled water as the control treatment.

Measured characteristics

The parameters evaluated at the end of the experiment (after three weeks) included vase life duration; percentage of opened, wilted and abscised florets; relative chlorophyll content using SPAD detection method measured on the first big leaf under inflorescence according to Coste et al. (2010); solution uptake; fresh weight; and relative water content (RWC) (Sairam et al. 2002). Before use, the SPAD was calibrated with an empirical relationship between SPAD unit and chlorophyll content by a homographic model. The homographic model was parameterized on the collected data set (N = 45 samples).

 $Chl_i = (\alpha \text{ SPAD}_i / (\beta - \text{ SPAD}_i)) + \varepsilon_i$

Chl_i is the total content of chlorophyll (a and b), α and β are parameters of the homographic models, and SPAD_i is the unitless reading from the SPAD-502 meter.

Experimental design and data analysis

Experiments were conducted in a complete randomized design with five replicates per treatment and two repetitions of the whole experiment in May and September 2015. Data were analyzed by SPSS-v.19 software with one-way analysis of variance test for CA and H_2O_2 separately. Furthermore, analysis of post-hoc Duncan test of general linear model univariate was used for input the comparisons effects of both biocides on postharvest of tuberose cut flower.

RESULTS

Data analysis showed significant effects of CA and H_2O_2 on the most measured characteristics of tuberose cut flowers. The only exceptions were, no effect of both compounds on fresh weight and H_2O_2 on the solution uptake (Table 1).

Table 1: F statistics from ANOVA for tested traits

Trait	Source of variation	
	Citric acid	H_2O_2
Solution uptake (ml)	3.911*	2.549 ns
Fresh weight (% of initial value)	1.172 ns	1.732 ns
Relative water content (%)	15.375**	7.742**
Wilting of florets (%)	6.437**	16.274**
Abscission of florets (%)	28.724**	29.755**
Chlorophyll content (mg·cm ⁻²)	43.564**	26.238**
Vase life duration (days)	12.841**	18.489**

ns – not significant, *p > 0.05, **p ≤ 0.05

Solution uptake, fresh weigh of cut flowers, and relative water content in leaves

CA at a concentration of 100 mg·dm⁻³ increased solution uptake by tuberose cut flowers ($F_{4,20} = 3.911$, P < 0.05), while H₂O₂ did not influence this trait (Table 1, Fig. 1). H₂O₂ and CA did not have a significant effect on fresh weight ($F_{4,20} = 1.172$ and $F_{4,20} = 1.732$, respectively) (Table 1, Fig. 2). The studied compounds significantly affected RWC of cut flowers during postharvest time [CA ($F_{4,20} = 15.375$, P < 0.01); H₂O₂ ($F_{4,20} = 7.742$, P < 0.01)] (Table 1). However, the effects differed between treatments (Fig. 3). The RWC increased at 50, 100, and 200 mg·dm⁻³ of CA and at 20 mg·dm⁻³ of H₂O₂.

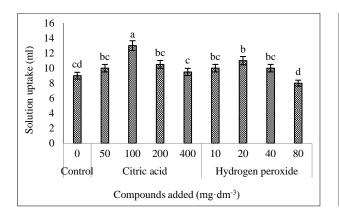


Fig. 1. Effects of citric acid and hydrogen peroxide on solution uptake by tuberose cut flowers. Bars indicating by the same letter do not differ significantly according to Duncan's multiple range test at p = 0.05, vertical lines – SE

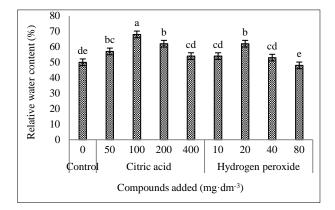


Fig. 3. Effects of citric acid and hydrogen peroxide on relative water content of tuberose cut flowers. Bars indicating by the same letter do not differ significantly according to Duncan's multiple range test at p = 0.05, vertical lines – SE

Percentage of the florets wilting and abscission

Application of CA and H₂O₂ significantly impacted the wilting of florets (CA: $F_{4,20} = 6.437$, P < 0.01; H_2O_2 : $F_{4,20} = 16.274$, P < 0.01, respectively) (Table 1). CA at all concentrations and H₂O₂ at 20, 40, and 80 mg dm⁻³ decreased flowers wilting (Fig. 4). At 200 mg dm⁻³ of CA, about 17% of florets wilted, and at 20 mg·dm⁻³ of H₂O₂, about 12% flowers wilted in comparison to 25% in the control without additions. The abscission of florets was retarded by the addition of CA and H₂O₂ at all conwith the greatest decrease centrations, at 100 mg \cdot dm⁻³ of CA, and at 20 and 40 mg \cdot dm⁻³ of H_2O_2 to about 15%, while in the control, about 33% of florets abscised (Fig. 5).

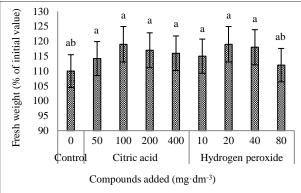


Fig. 2. Effects of citric acid and hydrogen peroxide on fresh weight of tuberose cut flowers. Bars indicating by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05, vertical lines – SE

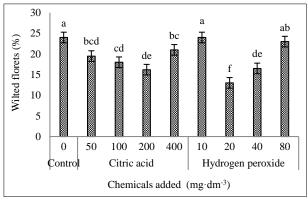


Fig. 4. Effects of citric acid and hydrogen peroxide on wilting of florets of tuberose cut flowers. Bars indicated by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05, vertical lines – SE

Fig. 5. Effects of citric acid and hydrogen peroxide on abscission of florets of tuberose cut flowers. Bars indicated by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05, vertical lines – SE

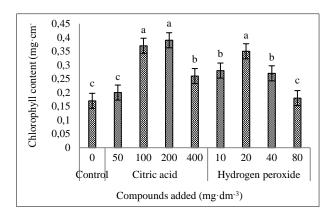


Fig. 6. Effects of citric acid and hydrogen peroxide on chlorophyll content of tuberose cut flowers. Bars indicated by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05, vertical lines – SE

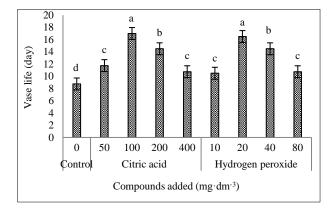


Fig. 7. Effects of citric acid and hydrogen peroxide on vase life of tuberose cut flowers. Bars indicated by the same letter do not differ significantly according to Duncan's multiple range test at P = 0.05, vertical lines – SE

Leaf chlorophyll content

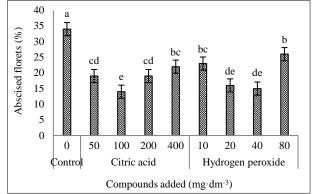
The compounds added to the water decreased chlorophyll loss significantly ($F_{4,20} = 43.564$, P < 0.01 for CA and $F_{4,20} = 26.238$, P < 0.01 for H₂O₂), but the effect depended on the kind of chemical and its concentration. Although all treatments of CA and H₂O₂ significantly maintained the leaf chlorophyll content in cut flowers in comparison with the control at the end of experiment after three weeks; the highest leaf chlorophyll content was obtained at 100 and 200 mg·dm⁻³ of CA and 20 mg·dm⁻³ of H₂O₂ (Fig. 6). At these concentrations, chlorophyll content was double compared to control.

Cut flowers vase life

Using CA and H_2O_2 as the additions to water during storage significantly increased the vase life in cut flowers of the tuberose ($F_{4,20} = 12.841$, P < 0.01 and $F_{4,20} = 18.489$, P < 0.01, respectively) (Table 1). Furthermore, these results showed that effects of CA and H_2O_2 on the cut flower vase life was dependent on concentration (Fig. 7). The longest vase life was obtained at 100 and 200 mg dm⁻³ CA (about 17 days) and 20 and 40 mg dm⁻³ H_2O_2 (about 17 and 15 days, respectively), while 9 days in the control.

DISCUSSION AND CONCLUSION

After cutting, the fresh weight of flowering stem decreases, which is caused by the reduction of water uptake and increasing water loss through evaporation and respiration (Borochov et al. 1995; Rattanawisalanon et al. 2003; Mansouri 2012; Liao et al. 2012). Our results showed the significant effects of 50 and 100 mg·dm⁻³ of CA and 20 mg·dm⁻³ of H₂O₂ in increasing the water uptake and nonsignificant effects on fresh weight of tuberose cut flowers during postharvest time. Our results are in accordance with previous reports showing the effects of low pH on inhibition of bacterial growth and prevention the early wilting in Dendrobium by Rattanawisalanon et al. (2003) and in chrysanthemums by Mansouri (2012). Adding CA to vase solution caused low latex flow from the cut stem surface and delay in the closure of xylem (Imsabai et al. 2013). Liao et al. (2012) reported that addition of H_2O_2 in



low concentration to vase solution improved the fresh weight of hybrid lily cut flowers and its antibacterial effect may be taken into account. Postharvest longevity of cut flowers is very important in their marketing and for economic value. Destruction of flowers by senescence is affected by some internal and external operants, such as genetics of cultivar, hormonal activity, polyamines, nutrient, and environmental factors (Leshem et al. 1998; Onozaki et al. 2001; Rogers 2006; Singh et al. 2008; Fanourakis et al. 2013; Teixeira da Silva et al. 2014; Perik et al. 2014). Furthermore, the browning and death of florets in cut flowers are the result of oxidative stress caused by polyphenoloxidase or peroxidases action that catalyze this process (Zhang et al. 2011). In tuberose, senescence symptoms are followed by browning and abscission of florets. The addition of CA and H_2O_2 to vase solution showed significant delay in browning and abscission of florets and, as a result, delaying senescence of tuberose cut flowers. Results showed that 100 mg dm⁻³ CA and 20 mg dm⁻³ H₂O₂ had the greatest effect in preventing browning, abscission, and senescence of florets, which approved the former studies where low levels of H₂O₂ was effective in delaying the senescence of lily cut flowers and rice leaves (Liao et al. 2012; Hung et al. 2006). In addition, several studies have shown that reduction in the pH of the vase solution decreases the senescence rate in cut flowers (Kazemi et al. 2011; Zhang et al. 2011; Perik et al. 2014).

Chlorophyll content is a qualitative marker of senescence of cut flowers at postharvest time (Podd & van Staden 2002; Mutui et al. 2006; Seglie et al. 2013; Daneshi Nergi & Ahmadi 2014). The results of the above studies showed a decrease in leaf chlorophyll content at postharvest time. According to findings of our research, delay in chlorophyll content loss improved the vase life of cut flowers. Both CA and H₂O₂ had a significant influence on delaying the chlorophyll loss during the tuberose postharvest time. The concentrations of CA - 100, 200, and $400 \text{ mg} \cdot \text{dm}^{-3}$ – and H_2O_2 – 10, 20, and 40 $\text{mg} \cdot \text{dm}^{-3}$ - induced a marked delay in decline of chlorophyll content, which is in agreement with other findings (Liao et al. 2012; Roberta Ansorena et al. 2014). A significant decrease in leaf RWC during vase life was obtained in the effect of the CA and H₂O₂

addition to the vase solution that inhibited the loss of water from leaves. Previous studies showed that H_2O_2 was involved in regulation of stomatal closure via abscisic acid activity and maintaining leaf water content (Zhang et al. 2001; Bright et al. 2006).

In the experiments described here, we have proven the positive effects of CA and H_2O_2 on morphological indices of tuberose cut flowers at postharvest. So, investigation on the physiological background is recommended for further studies.

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