

## The Role of Preservative Compounds on Number of Bacteria on the End of Stems and Vase Solutions of Cut Gerbera

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This study was conducted to evaluate the effect of preservative solutions on vase life, number of bacteria in the end of stem and in vase solution of cut gerbera 'Double Dutch' and 'Red Explotion'. Cut flowers were pulse-treated with nano-silver (2, 4, 6, 8 or 10 mg L<sup>-1</sup>) and thymol (12.5, 25, 50, 75 or 100 mg L<sup>-1</sup>) + 5 % sucrose. Experiment was conducted in completely randomized design with 5 replications and 1 flower in each experimental unit. Flower were harvested from a commercial greenhouse and transported to laboratory with 22±1°C temperature and 60±5% relative humidity. According to the results, these materials had positive effects on vase life of flowers. 6 mg L<sup>-1</sup> nano-silver treatments in 'Red Explotion' cultivar had highest longevity (14 days). All treatments were effective on decreasing of bacteria in stem end and solution. In 4 and 6 mg L<sup>-1</sup> SNP treatments was not any bacteria in vase solutions of 'Red Explotion' cultivar. Resulty showed that nano-silver can be use for increasing the vase life of cut gerbera 'Double Dutch' and 'Red Explotion'.

Abstract

**Keywords:** Anti microbial, Gerbera jamsonii, Nano-silver, Postharvaest, Thymol.

## INTRODUCTION

The colours of cut gerbera are very variable and gerbera is a popular cut flower (Syros *et al.*, 2004). Cut gerbera are sensitive to microbial contamination at the stem base (Balestra *et al.*, 2005). Many agents have been used in cut flower vase solutions to extend vase life by reduced microbial contamination. The bactericides are the most important components in the preservative solutions to control harmful bacteria and help to prevent bacterial embolism (Halevy and Mayak, 1981). Other material tested in preservative including sucrose (Kim and Lee, 2002) and thymol. Sucrose was used as substrate for respiration (Victoria *et al.*, 2003), it helps to maintain the osmotic potential of the petal cells (Sujatha *et al.*, 2003). Pre-treatment of cut roses with thymol have been found effective against some bacteria (Oraee *et al.*, 2010). Solgi *et al.*, (2009) reported that pre-treatment of *gerbera jamsonii* 'Dune' with nano-silver particles (SNPs) is important as an antibacterial agent (Morones *et al.*, 2005). Liu *et al.*, (2009) reported vase life extension for cut *gerbera jamsonii* 'Ruikou' flowers following pulsing with 5 mg L<sup>-1</sup> SNP solution for 24 h. Application of 10 mg L<sup>-1</sup> SNP + 5% sucrose for 24 h extended the vase life of cut *rosa hybrida* 'Dolce vita' flowers (Oraee *et al.*, 2010). Aim of present study is increasing vase life of cut gerbera by nano silver and thymol.

## MATERIALS AND METHODS

### Plant Material

Cut gerbera 'Double Dutch' and 'Red Explosion' were harvested from a hydroponic greenhouse in Mashhad, Iran. They were immediately stood upright in buckets partially filled with tap water and transported to the laboratory.

### SNP and Thymol Treatments

Flower stems were then re-cut under deionized water (DI) to 45 cm length. Cut gerberas were pulse-treated for 24 hours at 22±1 °C with 2, 4, 6, 8 or 10 mg L<sup>-1</sup> SNP (Nano cid company, Iran) and 12.5, 25, 50, 75 or 100 mg L<sup>-1</sup> thymol (Sigma company, USA) + 5% sucrose. Control flowers were pulse-treated with DI water. The flowers were kept under controlled conditions: 12 hours photoperiod at a photosynthetically activated radiation of 10 μmolm<sup>-2</sup> s<sup>-1</sup> provided by fluorescent lamps, 22±1 °C.

### Assessments of Vase Life

The end of vase life was defined the time that flowers were showing symptoms of petal wilting or curling and stem bending (Geraspolus and Chebli, 1999).

### Bacterial Counts

To determine the bacterial populations, vase water aliquots were tested during the experiment. Dilution carried out with 0.9% normal saline to achieve 30-300 bacterial colonies in one petri dish. 0.1 ml of aliquots was spread on nutrient agar plate. They were incubated at 37° C for 24 h before enumeration of bacteria (Bleeksma and Van Doorn, 2003).

To determine the stem bacteria numbers, 2 cm length segments were cut from the stem ends. Explants were washed three times with sterile DI to reduce the surface load of microbes. They were then grounded and diluted with 0.9% sterile normal saline. Liquid extract (0.1ml) was spread on nutrient agar plates and bacterial colonies were enumerated after incubation for 24h at 37° C (Balestra *et al.*, 2005).

### Statistical Analyses

Statistical significance between mean values was assessed using analysis of variance (ANOVA) and means were compared by Duncan's Multiple Range Test (DMRT; P≤ 0.05).

## RESULTS AND DISCUSSION

The results revealed that SNP and thymol at all concentrations increased vase life of all cultivars ('Double Dutch' and 'Red Explotion') compared to control. Our results were supported by some previous researches. They suggested that vase life of cut gerbera is greatly improved by chemical compounds and essential oils and there is a large difference among gerbera cultivars. Comparison of two cultivars showed that 'Red Explotion' had longevity more than 'Double Dutch'. There were significant differences between cultivars in response to SNP and thymol concentrations. Nano-silver at 6 mg L<sup>-1</sup> was the most effective treatment for increasing the vase life of gerbera jamsonii 'Double Dutch' (14 days). However, the SNP pulse treatment at 8 and 10 mg L<sup>-1</sup> caused visible damage to flowers. In both cultivars with increasing of thymol concentration from 12.5 to 100 mg L<sup>-1</sup>, the longevity of cut flowers increased (Table 1).

Our findings are similar to result of Solgi *et al.*, (2009), they reported that gerbera flowers are sensitive to microbial contamination and vase life of cut gerbera was extended by essential oils such as thymol and chemical compounds like SNP. Number of bacteria in the stem ends and vase solution tended to be increased throughout the gerbera vase life in the all treatments. There was significant difference in vase solution bacterial population between thymol and SNP pulse treatments. SNP reduced the number of bacteria and delayed the petal wilting. Pre-treatment with SNP (2, 4, 6 mg L<sup>-1</sup>) in both cultivars was generally very effective against bacterial growth, but 8 and 10 mg L<sup>-1</sup> concentrations had no effect on the number of bacteria and have toxic effect on cut gerbera. The highest number of bacteria was recorded in control ( $P \leq 0.05$ ). In 4 mg L<sup>-1</sup> and 6 mg L<sup>-1</sup> SNP in 'Red Explotion' was not any bacteria in the vase solution. In the other treatments, the number of bacteria was also less than control. There were significant differences in number of bacteria in the stem ends between the 100 mg L<sup>-1</sup> thymol and the control for the duration of assessment. In cut gerberas 'Double Dutch' and 'Red Explotion', we found that numbers of bacteria in stem end in third day was more than 106 CFU ml<sup>-1</sup> and in vase solution was less than 104 CFU ml<sup>-1</sup> (Table 1). Also there is relation between number of bacteria in vase solution and number of bacteria stem end (Oraee *et al.*, 2010). We concluded that all compounds had antibacterial effects and vase solution and stem ends microorganisms can decrease these preservative compounds. The most consistent antibacterial action was found in nano silver particles.

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## Tables

Table 1. Effect of different concentrations of nano silver particles and thymol on measured characteristics

Treatment		Vase life (Days)		No. of stem end bacteria in 3th day (CFU ml <sup>-1</sup> )		No. of vase solution bacteria in 3th day (CFU ml <sup>-1</sup> )	
<b>Cultivars</b>							
Control		6.2jkl*	4.4m	7.2×10 <sup>6</sup> d	8.9×10 <sup>6</sup> a	3.4×10 <sup>4</sup> c	8.4×10 <sup>4</sup> a
	2	9.2ef	6kl	5.6×10 <sup>4</sup> m	7.9×10 <sup>6</sup> c	8×10 <sup>3</sup> d	3.8×10 <sup>3</sup> h
	4	12.2b	9.6de	7.6×10 <sup>3</sup> t	5.1×10 <sup>4</sup> n	30j <	5.6×10 <sup>3</sup> f
SNP (mg L <sup>-1</sup> )	6	14a	10.6cd	5.1×10 <sup>2</sup> u	4×10 <sup>4</sup> q	30j <	8.4×10 <sup>2</sup> i
	8	11c	8gh	7.8×10 <sup>3</sup> s	4.9×10 <sup>5</sup> j	78j	3.2×10 <sup>3</sup> h
	10	10.4cd	7hijk	4.1×10 <sup>4</sup> p	8.4×10 <sup>5</sup> g	6.9×10 <sup>3</sup> e	8.3×10 <sup>3</sup> d
	12.5	7.2ghij	5.8l	8.05×10 <sup>3</sup> h	8.2×10 <sup>6</sup> b	4.1×10 <sup>3</sup> g	5.1×10 <sup>4</sup> b
	25	7.6ghi	6.4jkl	8×10 <sup>5</sup> i	7.2×10 <sup>6</sup> e	3.3×10 <sup>3</sup> h	3.3×10 <sup>3</sup> h
	50	9.2ef	6.8ijkl	5.6×10 <sup>4</sup> l	7.1×10 <sup>6</sup> f	8×10 <sup>3</sup> d	3.3×10 <sup>3</sup> h
Thymol (mg L <sup>-1</sup> )	75	10.2cde	7.6ghi	4.2×10 <sup>4</sup> o	8×10 <sup>5</sup> h	8.4×10 <sup>2</sup> i	3.3×10 <sup>3</sup> h
	100	11.2bc	8.2fg	7.9×10 <sup>3</sup> r	4.1×10 <sup>6</sup> k	7.1×10 <sup>2</sup> j	6.9×10 <sup>3</sup> e

\*Mean in each column with the same letters are not significantly different at 5% level of probability using DMRT.

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