

Research Article



Seed Halopriming Enhances Germination Performance and Seedling Vigor of *Gerbera jamesonii* and *Zinnia elegans*

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Abstract | A study was conducted to investigate the effects of seed priming (halopriming with 25 mM, 50 mM and 100 mM of calcium chloride (CaCl₂), potassium nitrate (KNO₃), potassium chloride (KCl) and Hydropriming) on germination and seedling vigor of *Gerbera jamesonii* and *Zinnia elegans*. Seeds of *Gerbera jamesonii* and *Zinnia elegans* were primed in salt solutions and distilled water for 24 h and 12 h, respectively, and germinated at 25 ± 1 °C with 40-60% R.H. in 16/8 hours day/night conditions. Results revealed that pre-sowing seed treatment particularly halopriming with CaCl₂ was the most effective for invigoration of gerbera and zinnia seeds. For *Gerbera jamesonii*, 25 mM CaCl₂ and for *Zinnia elegans*, 50 mM CaCl₂ proved best priming treatments followed by 100 mM KCl and 50 to 100 mM KNO₃ for both tested species. This was interpreted by reduced time to 50% germination (1.2 d for gerbera and 2.4 d for zinnia), higher final germination %age (11% for gerbera and 7% for zinnia), seedling vigor (83% for gerbera and 46% for zinnia), and fresh weight (0.95 g for gerbera and 2.29 g for zinnia) and dry weight (0.3 g for gerbera and 0.5 g for zinnia) of the seedlings. These halopriming treatments (25 to 50 mM CaCl₂ or 50 to 100 mM KCl and KNO₃) can be commercially used by the growers for early, uniform and healthy nursery stand of the tested species, which can ensure uniform maturity and flower harvesting of ornamentals.

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Introduction

Gerbera jamesonii, a perennial herb that flower during autumn and spring, is grown as an ornamental crop for specialty cut flower and bedding plant production (Dhingra et al., 2006). The elegant inflorescence is borne on a long stalk and the outermost petals (ray florets) may be creamy, reddish, yellow orange or pinkish in color (Bailly et al., 2003). *Zinnia elegans*, an annual flower of family Asteraceae, originated from Mexico, is a popular specialty cut

flower because of its large, colorful blooms and ability to withstand hot summers and is grown all over the world as specialty cut flower and bedding plant (Javid et al., 2005). Seed germination is an important stage in the life cycle of plants as it is directly involved in the effective seedling development, survival and population dynamics of specific crop. Bedding plant and cut flower producers pay deep attention to seed germination, because what is harvested is what initially emerges. Unfortunately, all thoughts of a uniform crop stand have long ago vanished due to thermo dor-

mancy and variation in seed vigor of seed lots, which make this goal difficult to achieve (McDonald, 2000). Production of specialty cut flowers has become a profitable business in many countries around the world and their popularity is increasing day by day. Gerbera and zinnia have some resistance to saline and other adverse environmental conditions (Gurusinghe et al., 2001). In recent years, a lot of work has been done on the invigoration of seeds to improve the germination rate and to reduce the emergence time of many crops (Basra et al., 2003). Several techniques employed to increase the seed germination with a purpose to shorten the emergence time and to protect seed from biotic and abiotic factors during critical phase of seedling establishment so as to synchronize emergence, which lead to uniform stand and improved yield.

Seed priming is a cheap tool to enhance the seedling emergence and establishment under marginal condition, as it reduces emergence time and provides better stand in many horticultural crops (Harris et al., 2001). Priming treatments known to enhance seed germination include hydropriming (Afzal et al., 2008), osmopriming (Afzal et al., 2006) and hormonal priming (Afzal et al., 2007). These techniques are helpful in controlling the hydration level within seeds so that the metabolic activity necessary for germination can occur but the emergence of radical is prevented which is most important step in priming technique (Afzal et al., 2002). Primed seeds germinate in a wider range of temperature (Esen et al., 2009) and are less sensitive to oxygen deprivation (Corbineau et al., 1993) than unprimed ones. The beneficial effect of priming has been associated with various biochemical, cellular and molecular events including synthesis of DNA and proteins (Bray et al., 1989). There is a serious problem in seed germination and uniform seedling growth of gerbera (Tjia, 1984; Cockshull, 1985; Moe et al., 1996) and uniform emergence of zinnia (Szopińska, 2011) greatly due to fluctuations in temperatures during nursery preparation (Farooq et al., 2004), which negatively affects synchronization of flowering in all plants, mandatory in landscape plants. Limited work has been reported so far regarding seed invigoration in gerbera and zinnia induced by seed priming. Keeping in view this scenario, this study was carried out to investigate the effect of different priming techniques for enhancing germination, uniform emergence and seedling vigor of gerbera and zinnia and to explore the possible basis of this enhancement.

Materials and Methods

This experiment was conducted to investigate the ef-

fect of different seed priming techniques on performance of *Gerbera jamesonii* and *Zinnia elegans* seeds under suboptimal temperature at Seed Physiology Laboratory, Department of Crop Physiology, University of Agriculture, Faisalabad, and under ambient temperature at Floriculture Laboratory, Institute of Horticultural Sciences, University of Agriculture, Faisalabad. The experiments were laid out according to completely randomized design individually for both species comprising eleven treatments viz. no priming, hydropriming, halopriming with 25, 50 or 100 mM of calcium chloride (CaCl_2), halopriming with 25, 50 or 100 mM of potassium chloride (KCl), or halopriming with 25, 50 or 100 mM of potassium nitrate (KNO_3) with each treatment replicated thrice. Seeds of *Gerbera jamesonii* and *Zinnia elegans* were obtained from a well-reputed private seed company. The ratio of seed and working solution was kept 1:5 (g mL^{-1}). Seeds of gerbera and zinnia were primed in aerated salt solutions of CaCl_2 , KCl or KNO_3 (halopriming; Applicam, Germany) or distilled water (hydropriming) for 24 h or 12 h, respectively. After treatment, seeds were washed thrice with distilled water and re-dried in oven near to their original weight. Seeds were then sealed in polythene bags and stored in a refrigerator at 5°C for later use.

Primed and non-primed (control) seeds (25 seeds per replicate of each treatment) were germinated on two folds of Whatman No. 1 filter paper, moistened with 4 mL distilled water, under 16/8 hours day/night conditions using fluorescent light at $25 \pm 1^\circ\text{C}$ and 40–60% R.H. for seven days. Data were recorded for time to 50% germination (T_{50} ; days) (Coolbear et al., 1984; Farooq et al., 2007), mean germination time (MGT; days) (Ellis and Roberts, 1981), final germination percentage (%; ratio of seeds germinated to total seeds tested), germination index (AOSA, 1990), root length (cm), shoot length (cm), root shoot ratio (root length/shoot length), seedling fresh weight (mg), seedling dry weight (mg), and seedling fresh and dry weight ratio (seedling fresh weight/seedling dry weight). Data were analyzed statistically according to analysis of variance (ANOVA) technique using Statistix 8.1 analytical software and treatment means were compared using Tukey's test (Steel et al., 1997).

Results and Discussion

Minimum mean germination time (MGT) (8.91 days) and time taken to 50% germination (3.25 days),

Table 1: Mean germination time, time to 50% germination and final germination percentage of gerbera and zinnia as influenced by various priming techniques.

Treatments	Mean germination time (days)		Time to 50% germination (days)		Final germination percentage (%)	
	Gerbera	Zinnia	Gerbera	Zinnia	Gerbera	Zinnia
Control (No priming)	9.94 a ^a	9.17 a	5.54 a	4.39 a	65.33 f	89.33
Hydropriming	9.59 b	8.67 b	4.93 ab	3.16 b	72.00 e	93.33
25 mM CaCl ₂	9.28 c	8.34 cd	4.32 bc	2.33 cde	76.00 cde	94.67
50 mM CaCl ₂	8.91 e	8.18 d	3.25 c	1.95 e	84.00 a	96.00
100 mM CaCl ₂	9.13 cd	8.34 cd	4.04 bc	2.16 de	78.67 bcd	96.00
25 mM KCl	9.25 c	8.65 b	4.21 bc	2.63 cd	73.33 e	94.67
50 mM KCl	9.11 d	8.40 c	3.95 bc	2.25 cde	78.67 bcd	93.33
100 mM KCl	9.11 d	8.30 cd	3.94 bc	2.00 e	81.33 ab	96.00
25 mM KNO ₃	9.21 cd	8.65 b	4.19 bc	3.18 b	76.00 cde	94.67
50 mM KNO ₃	9.10 d	8.38 c	3.32 c	2.12 e	80.00 abc	94.67
100 mM KNO ₃	9.16 cd	8.61 b	4.00 bc	2.67 c	74.67 de	93.33
Significance ^b	**	**	**	**	**	NS
LSD at P ≤ 0.05	0.17	0.20	1.07	0.4	1.33	NS

^a: Mean separation within columns by Tukey's test at P ≤ 0.05; ^b: P values were obtained using General Linear Models (GLM) procedures of Statistix 8.1 analytical software; ^{NS}: Nonsignificant at P > 0.05.

Table 2: Root length, shoot length and root-shoot ratio of gerbera and zinnia as influenced by various priming techniques.

Treatments	Root length (cm)		Shoot length (cm)		Root-shoot ratio	
	Gerbera	Zinnia	Gerbera	Zinnia	Gerbera	Zinnia
Control (No priming)	0.87 g ^a	4.67 e	0.70 d	3.17 e	1.31	1.49 abc
Hydropriming	1.20 f	6.33 bc	0.97 cd	4.00 bcde	1.24	1.59 abc
25 mM CaCl ₂	1.47 e	7.67 a	1.20 bc	5.67 a	1.22	1.35 c
50 mM CaCl ₂	2.00 a	6.33 bc	1.53 a	4.83 ab	1.31	1.31 c
100 mM CaCl ₂	1.77 b	5.50 cde	1.37 ab	3.83 bcde	1.30	1.44 bc
25 mM KCl	1.27 f	5.00 e	1.10 bc	3.50 cde	1.16	1.44 bc
50 mM KCl	1.50 cd	6.33 bc	1.23 bc	4.33 bcd	1.22	1.46 abc
100 mM KCl	1.67 c	7.17 ab	1.27 ab	4.83 ab	1.32	1.49 abc
25 mM KNO ₃	1.30 ef	6.00 cd	0.97 cd	3.33 de	1.35	1.80 a
50 mM KNO ₃	1.63 bcd	7.17 b	1.30 ab	4.17 bcde	1.26	1.73 ab
100 mM KNO ₃	1.50 cd	7.67 a	1.17 bc	4.50 bc	1.28	1.72 ab
Significance ^b	**	**	**	**	NS	**
LSD at P ≤ 0.05	0.13	0.33	0.23	0.17	NS	0.07

^a: Mean separation within columns by Tukey's test at P ≤ 0.05; ^b: P values were obtained using General Linear Models (GLM) procedures of Statistix 8.1 analytical software; ^{NS}: Nonsignificant at P > 0.05.

while maximum final germination percentage (FGP) (84.0%) of gerbera seeds were recorded when seeds were primed using 50 mM CaCl₂ (Table 1). Maximum root length (2.0 cm) and shoot length (1.53 cm) was recorded by using 50 mM CaCl₂ as priming agent (Table 2). Root-shoot ratio was similar for all priming treatments ranging from 1.16 to 1.35 and averaged 1.27 (Table 2). Maximum fresh (3.91 g) and dry weight (1.15 g) of gerbera seedlings were record-

ed when seeds were primed using 50 mM CaCl₂ (Table 3). While fresh and dry weight ratio was similar for all priming treatments which averaged 3.33.

For zinnia, mean germination time (8.18 days) and time to reach 50% germination (1.95 days) was reduced due to seed treatment with 50 mM CaCl₂. Final germination percentage of zinnia was statistically similar for all priming agents and averaged

Table 3: Fresh weight, dry weight and fresh-dry weight ratio of gerbera and zinnia as influenced by various priming techniques.

Treatments	Fresh weight (g)		Dry weight (g)		Fresh-dry weight ratio	
	Gerbera	Zinnia	Gerbera	Zinnia	Gerbera	Zinnia
Control (No priming)	2.18 g ^a	3.27 e	0.66 f	1.20 c	3.30	2.86
Hydropriming	2.88 f	3.97 de	0.87 e	1.19 c	3.31	3.36
25 mM CaCl ₂	3.13 de	4.66 cd	0.95 d	1.40 abc	3.30	3.32
50 mM CaCl ₂	3.91 a	5.56 a	1.15 a	1.67 a	3.41	3.34
100 mM CaCl ₂	3.49 c	5.01 ab	1.05 bc	1.51 bc	3.33	3.33
25 mM KCl	3.54 c	4.98 abc	1.06 bc	1.50 c	3.33	3.33
50 mM KCl	3.56 c	5.47 a	1.07 bc	1.64 b	3.34	3.35
100 mM KCl	3.76 ab	5.29 ab	1.13 ab	1.58 ab	3.33	3.35
25 mM KNO ₃	2.99 ef	4.30 d	0.89 e	1.29 bc	3.37	3.33
50 mM KNO ₃	3.22 d	4.71 bc	0.96 d	1.42 abc	3.35	3.33
100 mM KNO ₃	3.65 bc	5.23 ab	1.13 b	1.58 ab	3.26	3.31
Significance ^b	**	**	**	**	NS	NS
LSD at P ≤ 0.05	0.09	0.18	0.02	0.03	NS	NS

^a: Mean separation within columns by Tukey's test at P ≤ 0.05; ^b: P values were obtained using General Linear Models (GLM) procedures of Statistix 8.1 analytical software; NS: Nonsignificant at P > 0.05.

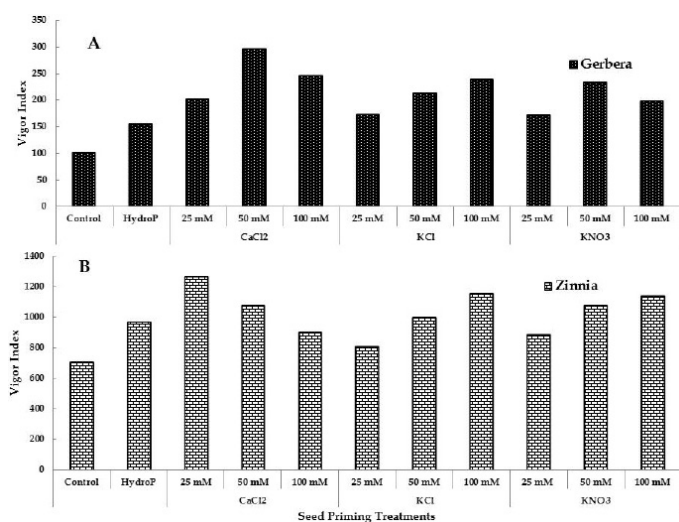


Figure 1: Vigor index of gerbera (A) and zinnia (B) seedlings as influenced by various seed priming treatments.

94.18%; maximum (96.00%) in 100 mM CaCl₂ and minimum (89.33%) in non-primed seeds. Maximum shoot (7.67 cm) and root length (5.67 cm) of zinnia seedlings were recorded in priming treatments comprising 25 mM CaCl₂. Seed priming with 25 mM KNO₃ had highest root-shoot ratio of zinnia (1.80). Maximum fresh (5.56 g) and dry weight (1.67 g) of zinnia seedlings were recorded in priming treatment comprising 50 mM CaCl₂, while no significant difference was noticed for fresh and dry weight ratio that ranged from 2.86 for non-primed (control) seeds to 3.36 for hydroprimed seeds and averaged 3.29. It is evident from results (Table 1) that 100 mM KCl and 50 mM KNO₃ reduced T₅₀ and were statistically sim-

ilar to 50 mM CaCl₂. Moreover, these treatments also decreased MGT and increased FGP for both species. Use of 50 mM CaCl₂ improved fresh and dry weight of both tested species.

Germination and seedling establishment are critical stages in the life cycle of plants especially under stress conditions. There is a strong evidence available that pre-sowing treatments with inorganic salts not only promotes seed germination but also enhance later growth, metabolic processes, and, hence, ultimate increase crop yield (Cantliffe, 1998). Seed priming has proven beneficial in this regard in many important horticultural crops (Khan et al., 2012; Batool et al., 2015). Seed priming showed significantly better results over control for time taken to 50% germination, final germination percentage, root and shoot length, seedling fresh and dry weight and seedling vigor (Tables 1, 2 and 3; Figure 1) (Afzal et al., 2009; Mukhtar et al., 2013). Primed seeds significantly exhibited higher germination percentage, speed of germination, root and shoot length and seed vigor in both gerbera and zinnia. Yoon et al. (1997) reported that pansy seeds primed with CaCl₂ showed significant reduction in time of emergence. Germination enhancement due to priming has been attributed to membrane repair process, mobilization of storage reserves towards growing points, advancement of germination to end of phase II or osmotic adjustments during priming (Bradford, 1986). Our findings are also in line with

the results of Demir and Oztokat (2003), who reported that seed priming using salts decreased the mean germination time in watermelon.

Halopriming plays a primary role in regulating the activities of existing enzymes, thereby producing germination metabolites in requisite amounts. Shoot and root length was increased due to halopriming with CaCl_2 over the control, which might be due to higher cell wall extensibility by the primed seeds as observed in marigold (Afzal et al., 2009) and rice (Hussain et al., 2017). In this study, CaCl_2 significantly increased root and shoot length possibly due to extensibility of cell wall in roots because Ca^{+2} have a fundamental importance for the maintenance of root elongation in seedlings (Ghiyasi et al., 2008). The activation of membrane repair due to priming (Khan et al., 2016) might have reduced electrical conductivity of seed leachates, which might have resulted in higher reducing and total sugars as well as higher α -amylase activity in haloprimed seeds (Smith and Cobb, 1991). Ca^{+2} acts as a second messenger in plant cells, being involved in the mediation of signaling pathways (Hussain et al., 2006) and might have evoked genes related to antioxidants or antioxidant enzymes.

Conclusion and Recommendations

It is concluded that priming with 50 mM CaCl_2 followed by 100 mM KCl and 50 mM KNO_3 enhanced uniform and improved seed germination and thus improved vigor of both tested species, which may be used by the growers/nurserymen for uniform and best quality flower production.

Authors' Contribution

All co-authors equally contributed in the manuscript. The principal author, Iftikhar Ahmad, was involved in planning of the study, its execution, write-up of the manuscript and submission. Abdul Manan Saleem was responsible for data collection and helping in write up of the paper. Ghulam Mustafa was involved in execution of the experiment, data collection, taking care of plants and data analysis. Khurram Ziaf was involved in application of treatments and editing manuscript. Irfan Afzal was responsible for planning and application of treatments and Muhammad Qasim edited the manuscript.

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