

# Improvement of eggplant seed germination and seedling emergence at low temperature by seed priming with incorporation SA into KNO<sub>3</sub> solution

Yanping ZHANG<sup>1,2</sup>, Haihe LIU<sup>1,2</sup>, Shuxing SHEN (✉)<sup>1</sup>, Xine ZHANG<sup>1</sup>

<sup>1</sup> College of Horticulture, Agricultural University of Hebei, Baoding 071001, China

<sup>2</sup> College of Agronomy, Engineering University of Hebei, Handan 057150, China

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**Abstract** The effects of incorporation SA into KNO<sub>3</sub> priming solution on the germination and emergence of eggplant seeds at 15°C were investigated. Seeds were primed into 3% KNO<sub>3</sub> containing 0.05, 0.1, 0.5, or 1 mM salicylic acid (SA) for 6 days, respectively. After the priming, seeds were either immediately used for germination and emergence test at 15°C or stored at 4°C for 1 month and then for the germination test. The primed eggplant seeds in general improved the final germination percentage (FGP), germination rate (G<sub>50</sub>), and germination synchrony (E<sub>10–90</sub>) at 15°C compared with non-priming seeds. Priming seeds in 3% KNO<sub>3</sub> solution supplement with 0.1 mM SA resulted in the best priming effect compared with other priming treatments and non-priming treatment, and all of the beneficial priming effects were still retained after stored at 4°C for 30 days. These results indicated that priming seeds in 3% KNO<sub>3</sub> solution containing 0.1 mM SA could be used as an effective method to improve low-temperature performance of eggplant seeds and subsequent seedling growth.

**Keywords** eggplant (*Solanum melongena* L.), priming, germination, low temperature, SA, KNO<sub>3</sub>

## Introduction

Eggplant is a species that requires a relatively high soil temperature for germination and emergence, and the optimal germination temperature ranges from 25°C to 30°C. When eggplant seeds are sowed directly in the field in winter or early spring in north of China, the soil temperature is often lower than the optimal temperature, causing delayed or non-uniform seedling emergence. Failure of obtaining fast and uniform emergency is a major limiting factor for eggplant production.

Low temperature is one of the most important environmental factors affecting eggplant seed germination or seedling emergence. The incorporation of plant growth regulators into the priming solution has been shown to be effective to enhance germination and seedling emergence of crops under adverse condition. For example, imbibing tomato

and bean seeds in acetyl salicylic acid (ASA) solution increased seedling survival during subsequent chilling, high temperature, and drought stress (Tissa et al., 2000), while incorporation of ASA or methyl jasmonate (MeJA) into the priming solution improved sweet pepper germination and emergence at low temperature (Ahmet, 2005). However, most of the eggplant seed priming studies have been focused on improving germination under normal condition (Wu et al., 2001; Li et al., 2006; Yao and Song, 2006; Chen et al., 2008), while little information is available on improving the performance of eggplant seeds at low temperature. The objectives of this study were to investigate effects of priming with incorporation of salicylic acid (SA) into priming solution on subsequent germination and seedling emergence at low temperature and confirm if the effects could be retained after priming and stored for 30 days.

## Materials and methods

### Materials and pretreatment

The seeds of eggplant cv. ‘Kuaiyuan’ used in this study were

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Correspondence: Shuxing SHEN

E-mail: shensx@hebau.edu.cn

produced by Tianjin Vegetable Research Institute, China. The initial germination percentage was 96% at normal temperature. SA was purchased from Beijing Chemical Agents Co. All experiments reported in this study were carried out in the beginning of 2011 at the College of Agronomy, Hebei Engineering University, China. Seeds were disinfested in 1% sodium hypochlorite for 15 min to eliminate seed-borne pathogens, rinsed under running tap water for 1 min, and dried by spreading them on absorbent paper for 30 min under room condition at 20°C and 50% RH.

## Methods

### *Seed priming*

Seed priming procedures consisted of soaking 5 g eggplant seeds for 6 days at 25°C in 3% KNO<sub>3</sub> solution containing 0.05, 0.01, 0.5, or 1.0 mM SA, respectively, and changing the priming solution supplemented with SA everyday to maintain a constant osmotic potential. Seeds priming in 3% KNO<sub>3</sub> solution containing no SA and non-primed seeds were used as controls in the experiment. Thereafter, the seeds from each priming solution were rinsed under tap water for 1 min and dried on absorbent paper under room condition at 20°C and 50% RH for 2 h. Seed germination and seedling emergence were immediately evaluated after priming.

### *Seed germination test at low temperature*

Germination test was carried out in a temperature-controlled incubator at 15°C in darkness; primed and non-primed seeds were respectively spread on two layers of filter paper moistened with 5 mL distilled water in covered 9 cm diameter Petri dishes. Treatments were arranged in a completely randomized design with four replications of 50 seeds. Radical protrusion to 1 mm was scored as germination. Germination was recorded daily for 14 days and germinated seeds were removed from the Petri dishes. From the total number of seed germinated, the final germination percentage (FGP), its angular transformation ( $\text{degree} = \arcsine\sqrt{\text{FGP}}$ ), days used to reach 50% FGP ( $G_{50}$ ), as well as days between 10% and 90% of FGP ( $G_{10-90}$ ) were calculated.

### *Seedling emergence test at low temperature*

Seeds were primed as described above and 20 seeds from each treatment were planted at 1.5 cm depth into a 50-cell plastic cell tray filled with growth medium consisting of peat and vermiculite in a ratio of 2:1. The cell tray was watered and placed in an incubator at 15°C under a 14 h/d light period at seeding stage. Each treatment was replicated four times and arranged with a randomized complete block design in the cell tray. Emergence counts were recorded daily, and final emergence percentage (FEP) and its angular transformation ( $\text{degree} = \arcsine\sqrt{\text{FEP}}$ ), days used to reach 50% FEP, and days between 10% and 90% of FEP were calculated. Twenty-five days after planting, when the

percentage of emergence had stabilized in all treatments, the number of seedlings that emerged and lived, shoot fresh (cut at the medium surface) and dry weights (dried at 65°C for 48 h) were recorded.

### *Seed germination at low temperature following storage*

Seeds were primed as described above. Because 0.5 and 1.0 mM SA acid added to the priming solution significantly decreased the germination rate of eggplant seeds at 15°C, both concentrations were not included in the seed storage experiment. Following the priming, seeds were rinsed as described above and dried by spread on absorbent paper for 2 days under room conditions at 20°C and 50% RH. Non-primed seed were also subjected to the same drying condition. Primed and non-primed seeds then were severally sealed in a small plastic bag and sorted at 4°C for 30 days. At the end of storage, seeds were subjected to germination test at 15°C as described above.

### *Statistical analysis*

Analysis of variance was carried out with the data from all experiments. When data expressed as percent, an arcsine transformation was used prior to analysis of variance. Means separation was performed by least significant difference (LSD) test if *F*-test was significant at  $P = 0.05$ .

## Results

### **Seed germination at low temperature**

Priming eggplant seeds, whether SA added to the priming solution or not, significantly improved germination percentage at 15°C compared with non-priming seeds which had a FGP of 41% (Table 1). Incorporation of 0.1 mM SA into the priming solution resulted in the highest of FGP (92%). However, with the increase of the concentration of SA from 0.1 to 1.0 mM, FGP of eggplant seeds was significantly reduced from 92% to 64%. All priming treatments also significantly improved the eggplant seed germination rates compared with the non-priming seeds, which reached 50% FGP ( $G_{50}$ ) in 8.1 days, and the minimum  $G_{50}$  was also obtained from seeds primed in KNO<sub>3</sub> supplemented with 0.1 mM SA ( $G_{50} = 2.4$  days). Among the priming treatments, the priming treatments in the presence of 0.05 mM SA and 0.1 mM SA improved the germination synchrony compared with non-priming treatment. For both of them, the days between 10% and 90% of FGP ( $G_{10-90}$ ) were 6.5 and 6.3 days, respectively.

### **Seedling emergence at low temperature**

Seed priming significantly increased the FEP of eggplant seedlings at low temperature compared with non-priming seeds (Table 2). The effects of KNO<sub>3</sub> in the presence of SA

**Table 1** Results of eggplant seed germination in darkness at 15°C primed in 3% KNO<sub>3</sub> with various concentrations of SA for 6 days at 25°C

| Treatments                    | FGP |          | G <sub>50</sub> (days) | G <sub>10-90</sub> (days) |
|-------------------------------|-----|----------|------------------------|---------------------------|
|                               | (%) | (degree) |                        |                           |
| KNO <sub>3</sub> + 0.05 mM SA | 84  | 67       | 5.1                    | 6.5                       |
| KNO <sub>3</sub> + 0.1 mM SA  | 92  | 74       | 2.4                    | 6.3                       |
| KNO <sub>3</sub> + 0.5 mM SA  | 76  | 61       | 5.9                    | 7.4                       |
| KNO <sub>3</sub> + 1 mM SA    | 64  | 54       | 6.6                    | 7.8                       |
| KNO <sub>3</sub> + 0 mM SA    | 58  | 50       | 5.4                    | 6.9                       |
| Non-priming seeds             | 41  | 40       | 8.1                    | 7.5                       |
| LSD <sub>0.05</sub>           | —   | 7.8      | 0.7                    | 0.7                       |
| Significance                  | —   | S        | S                      | S                         |

S means significant difference at  $P \leq 0.05$ .

**Table 2** Results of eggplant seed emergence at 15°C primed in 3% KNO<sub>3</sub> with various concentrations of SA for 6 days at 25°C

| Treatments                    | FEP |          | E <sub>50</sub> (days) | E <sub>10-90</sub> (days) | Survival (%) | Shoot fresh wt. (mg per plant) | Shoot dry wt. (mg per plant) |
|-------------------------------|-----|----------|------------------------|---------------------------|--------------|--------------------------------|------------------------------|
|                               | (%) | (degree) |                        |                           |              |                                |                              |
| KNO <sub>3</sub> + 0.05 mM SA | 78  | 63       | 16.4                   | 6.7                       | 96           | 34.8                           | 4.4                          |
| KNO <sub>3</sub> + 0.1 mM SA  | 86  | 69       | 16.2                   | 6.3                       | 95           | 36.4                           | 4.6                          |
| KNO <sub>3</sub> + 0.5 mM SA  | 72  | 59       | 17.6                   | 8.9                       | 92           | 31.2                           | 4.0                          |
| KNO <sub>3</sub> + 1 mM SA    | 50  | 46       | 18.2                   | 8.8                       | 93           | 30.6                           | 3.7                          |
| KNO <sub>3</sub> + 0 mM SA    | 53  | 47       | 17.8                   | 8.1                       | 92           | 31.8                           | 3.8                          |
| Non-priming seeds             | 41  | 39       | 21.2                   | 7.2                       | 95           | 24.2                           | 3.1                          |
| LSD <sub>0.05</sub>           | —   | 5.8      | 1.4                    | 0.8                       | —            | 4.6                            | 0.8                          |
| Significant                   | —   | S        | S                      | S                         | NS           | S                              | S                            |

NS means non-significant difference at  $P \leq 0.05$ ; S means significant difference at  $P \leq 0.05$ .

were generally more effective than KNO<sub>3</sub> alone. Furthermore, the effects of KNO<sub>3</sub> added 0.05 or 0.1 mM SA was significantly better than KNO<sub>3</sub> alone for FEP and E<sub>50</sub>. The seeds primed in the presence of 0.1 mM SA resulted in the highest FEP (86%) and the least E<sub>50</sub> (16.2 days). The emergence synchrony was significantly improved only by priming solution containing 0.05 mM SA. Compared with lower concentration of SA ( $\leq 0.1$  mM), the higher concentration of SA ( $> 1$  mM) significantly reduced the emergence percentage and rate of eggplant seedlings at low temperature.

All treatments had  $> 92\%$  survival rates and there were no significant difference among the treatments. Both fresh and dry shoot weights of eggplant seedling were significantly affected by priming treatments, and the priming in the presence of 0.1 mM SA resulted in the highest fresh and dry shoot weights.

#### Seed germination at low temperature following storage

After 30 days of storage, all priming treatments still significantly improved FGP and germination rate of eggplant seeds at low temperature compared with non-priming seed (Table 3). The results indicated that beneficial effects of the seed priming with the inclusion of SA were retained within 30 days storage at 4°C. Priming in the presence of 0.1 mM SA resulted in the highest FGP (84%) and the fast germination

rate (G<sub>50</sub> = 4.1 days). The germination synchronies of primed seeds were significantly improved compared with non-priming seeds, but there was no significant difference among the priming treatments.

## Discussion

The beneficial effects of seed priming with inclusion of plant growth regulator into priming solution to improve germination and emergence at low temperature have been reported on some crop species, such as muskmelon (Ahmet et al., 2004), sweet pepper (Ahmet, 2005), cucumber (Shu et al., 2006; Yang et al., 2007), and sugar beet (Mostafa et al., 2007). The results of this study on eggplant were very consistent with the above reports and indicated that the priming eggplant seeds with incorporation 0.1 mM SA into 3% KNO<sub>3</sub> resulted in better germination and emergence at 15°C compared with the non-priming and priming in KNO<sub>3</sub> solution alone.

Increasing evidences suggested that benzoic acid derivatives such as SA or ASA regulated the stress tolerance in plants. These molecules triggered the expression of the potential to tolerate stress rather than having any direct effect as a protectant (Yin and Hou, 2007). Mendoza et al. (2002) reported that imbibing pepper seeds in 0.1 mM SA prevented seedling from subsequent chilling-induced damage. Tissa et al. (2003) founded that soaking bean and tomato seeds in

**Table 3** Results of eggplant seed germination in darkness at 15°C primed in 3% KNO<sub>3</sub> with various concentrations of SA for 6 days at 25°C and stored at 4°C for 30 days

| Treatments                    | FGP |          | G <sub>50</sub> (days) | G <sub>10-90</sub> (days) |
|-------------------------------|-----|----------|------------------------|---------------------------|
|                               | (%) | (degree) |                        |                           |
| KNO <sub>3</sub> + 0.05 mM SA | 82  | 66       | 4.8                    | 6.2                       |
| KNO <sub>3</sub> + 0.1 mM SA  | 84  | 67       | 4.1                    | 6.4                       |
| KNO <sub>3</sub> + 0 mM SA    | 58  | 50       | 5.6                    | 6.8                       |
| Non-priming seeds             | 48  | 44       | 7.8                    | 7.3                       |
| LSD <sub>0.05</sub>           | —   | 5.2      | 1.2                    | 0.8                       |
| Significant                   | —   | S        | S                      | S                         |

S means significant difference at  $P \leq 0.05$ .

0.1 mM ASA for 24 h resulted in 100% seedling survival following chilling and heat stresses, while none of the control plants survived. The fact that seeds imbibing with ASA or SA provided multiple stress tolerance in tomato and bean plants was more consistent with a signaling role for the expression of tolerance rather than a direct effect.

In seed priming studies, the primed seeds that immediately performed germination test resulted in very rapid germination, while the storing primed seeds generally reduced germination and emergence. To retain the beneficial effects of primed seed for a long time, the primed seeds should be stored at low temperature and low moisture. In this study, although there was a slight reduction in germination percentage and rate of primed eggplant seeds stored for 30 days compared with their performance before the storage, seeds from all primed treatments performed better than non-priming ones. High germination percentage and fast germination indicated that beneficial effects of primed seed were retained within 30 days of storage at 4°C.

In summary, this study demonstrated that seed priming with the inclusion of 0.1 mM SA into 3% KNO<sub>3</sub> solution significantly improved low temperature germination and emergence of eggplant seeds at low temperature (15°C) compared with the seeds primed in KNO<sub>3</sub> solution alone and non-primed seeds. The result indicated that SA into KNO<sub>3</sub> solution could serve as an effective way to improve seeds germination and seedlings emergence of eggplant at low temperature. Moreover, the study also revealed that the beneficial effects of seeds priming were still retained within 30 days of storage at 4°C.

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