PRIMING ON EMBRYO EMERGENCE AND SEEDLING VIGOR OF SMALL FRUITED BITTER GOURD (MOMORDICA CHARANTIA L.) UNDER SUB-OPTIMAL TEMPERATURE

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ABSTRACT

An experiment was conducted to observe the effect of priming on seedling emergence and vigor under sub-optimum temperature. Six treatments namely: T1- control (no soaking), T2- tap water soaking for 24 hours, T3- warm water soaking at 50°C for 60 minutes, T4- 1% KNO3 solution soaking at 25°C for 48 hours, T5- 10 mgL⁻¹ sodium selenite soaking at 25°C for 48 hours, T6- 400 mgL⁻¹ GA3 solution soaking for 24 hours were imposed in the experiment. The results indicated that priming significantly improved seedling emergence at low temperature (20°C). Both emergence and speed of germination were increased in priming treatments especially in warm water treated seeds. Primed seeds at sub-optimum temperature enhanced emergence percentage at a faster rate. Thus, more than 90% of primed seeds emerged within 8 days against 14 days required for non-primed seeds. The enhanced germinability has been related to priming-induced quantitative change in biochemical content of the seeds and membrane integrity and to enhance physiological activities at seeds germination. However, cumulative emergence rate varied widely among the primed and non-primed seeds. The higher percent emergence and faster rate of emergence was obtained by treating seeds with GA3 solution. Priming treatments improved percent emergence by 95, 93 and 89 % in both of GA3 and warm water, KNO3 and sodium selenite, respectively. It was found from the study that priming treatment significantly increased vigor index. The emergence index was significantly influenced by priming of seeds which varied from 33.60 to 12.14. Among the priming treatments, GA3 produced more vigorous seedlings and the highest shoot length and shoot mass.

KEY WORDS: Bitter gourd, seed priming, seedling vigor, temperature

INTRODUCTION

Bitter gourd is a tropical vine preferring high humidity and warm weather. For successful seedling emergence, it requires temperature range from 25-28°C. Germination ceases outside the range of 10-50°C. It may fail or take a long time to germinate if the soil temperature is too low. In addition, bitter gourd seeds rapidly lose viability and its thick seed coat enclosing embryo affect germination by imposing mechanical restriction on embryo growth.
Slow and poor emergence is a common problem of this vegetable cultivation due to sub-optimum temperature (<20°C) prevailing in winter. Normally, seedlings emerge 6-8 days after sowing (Reyes et al., 1994). While, seeds sown directly in the open field fail to germinate or germination is delayed remarkably (Joshi and Srivastava, 2002). The aging reduced seed germination enhanced lipid peroxidation and decreased anti-oxidative responses of bitter gourd seeds. Priming the seeds prior to aging treatment partially overcame the deteriorative effect of aging and improved the quality of seed by decreasing lipid peroxidation and enhancing anti-oxidative responses.

Seed priming, one of the techniques to promote germination, advances the physiological status of seeds just before root extrusion by controlling the water supply or other priming agents. On the other hand, priming offers an effective means for counteracting sub-optimum temperature induced oxidative injury and raising seed performance in many crop species (Chen and Sung, 2001). However, priming is a technique in which seeds are partially hydrated so that the pre-germinative metabolic activities are initiated, but radical emergence does not occur (Bradford, 1986, and Pill, 1986). Bradford (1986) further suggested that a good measure of the effectiveness of priming might be the ability of seed to germinate at reduced water potential or under adverse temperatures. It is an effective technique for rapid and uniform germination of several crops (Basra et al., 2005 and Farooq et al., 2005). Application of plant growth regulators induce greater breakdown of seed reserves in storage tissue and/or increase the activity of enzymes concerned with mobilization resulted in improved seed germination (Bewley and Black, 1986).

For enhancing germination and improved stand establishment, seed priming has been suggested (Harris, 1996). Seeds treated with osmotic solutions ranging from -1.0 to -2.0 mPa water potential may germinate more rapidly and uniformly under a wide range of temperature than untreated seeds (Pessarakli, 2002). Osmotic salt solutions for priming are KNO$_3$, KCl, K$_2$HPO$_4$, KH$_2$PO$_4$, MgSO$_4$, CaCl$_2$, and NaCl used to control water uptake and prevent radical protrusion, and salt also supply N and other nutrients needed for protein synthesis (Bray, 1995). Therefore, in view of the above facts, the present investigation was conducted to evaluate the effect of priming practices on germination capacity and seedling vigor of bitter gourd, to alleviate the problem of low and non-synchronous emergence of bitter gourd seedling.

MATERIALS AND METHODS

The experiment was conducted at the laboratory and research farm of Crop Botany Department, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Bangladesh. Locally collected small type bitter gourd landrace BG-5 was used as test material. The experiment was laid out in a completely randomized block design with three replications. Six treatments namely: T$_1$, control (no soaking), T$_2$, tap water soaking for 24 hours, T$_3$, warm water soaking at 50°C for 60 minutes, T$_4$, 1% KNO$_3$ solution soaking at 25°C for 48 hours, T$_5$, 10 mgL$^{-1}$ sodium selenite soaking at 25°C for 48 hours, T$_6$, 400 mgL$^{-1}$ GA$_3$ solution soaking for 24 hours. Fifty seeds of BG-5 for each treatment were taken for germination, and were soaked by tap water for 24 h (T$_2$) and warm water soaking was done by soaking the seeds in water at 50°C for 60 minutes (T$_3$) and then air dried them at room temperature. Thereafter, seeds were soaked in 1% KNO$_3$ solution (T$_4$) and 10 mgL$^{-1}$ sodium selenite (T$_5$), and seeds were
incubated at 25°C for 48 h. After the completion of incubation period, seeds were washed in running tap water and then air drying them at room temperature. Seeds were also soaked in GA3 (400ppm) solution for 24 h (T6) and then air-drying them at room temperature. While, seeds of control treatment (T1) were not soaked i.e. seeds were directly sown in polyethylene bags. After soaking, the air-dried seeds were used for germination using three replicates. Fifty seeds were planted in polyethylene bags which were filled with cow dung mixed soil (1:2). These polyethylene bags were watered as and when required. Number of seedlings was counted daily until no further emergence occurred for 12 days after sowing (DAS) and percent emergence was calculated using the following formula:

\[
\% \text{ Emergence} = \frac{\text{Number of emerged seedlings}}{\text{Number of seeds sown}} \times 100
\]

**Speed of Germination**

Copeland (1976) considered both vigor index and co-efficient of germination as measures for speed of germination. Counting the germinated seedlings at an interval of 24 hours for 8 days and the speed of germination of seed was monitored. Vigor index, Co-efficient of germination and germination index were calculated using following formula (Copeland, 1976):

\[
\text{Vigor index} = \frac{A_1}{T_1} + \frac{A_2}{T_2} + \frac{A_3}{T_3} + \ldots + \frac{A_x}{T_x}
\]

Where,

\[
A = \text{number of seed germinated}
\]

\[
T = \text{number of days corresponding to } A
\]

\[
x = \text{counting number (1, 2, 3, \ldots nth )}
\]

\[
\text{Co efficient of emergence (%) } = \frac{100(A_1 + A_2 + A_3 + \ldots + A_x)}{A_1T_1 + A_2T_2 + \ldots + A_xT_x} \times 100
\]
Where, \( A \) = number of seed germinated
\( T \) = time corresponding to \( A \)
\( x \) = counting number (1, 2, 3, .............\( n \)th)

\[
\text{Emergence index} = \frac{\sum \text{Ti Ni}}{S}
\]

Where, \( Ti \) = \( i \)th number of days after sowing
\( Ni \) = \( i \)th number of seeds emergence
\( S \) = total no. of seed used

Shoot and Root Length

Plumule and radicle length were measured after 30 days of sowing, ten seedlings per replicate were harvested and the shoot and root length of individual seedlings was recorded manually. The seedlings were separated into root, shoot and were dried at 70°C for 72 hours in an electric oven and the weight was recorded.

Statistical Analysis

The data were analyzed by portioning the total variance with the help of computer using MSTAT program. The treatment means were compared using LSD at 5% level of significant.

RESULTS

Seedling Emergence Percentage

Seedlings emerged protruding soil surface was considered as emergence. Fig. 1 showed cumulative emergence of ‘uchja’ seedlings for a period of 12 days following seed sowing. Seed priming increased percent emergence and reduced time to reach the maximum percent emergence (Fig. 1). It is evident from the results in Fig. 1 that the priming treatment had significant positive influence on the seedling emergence. The rate of emergence and eventually the cumulative seedling emergence were much lower in control treatment than the other five treatments. \( \text{GA}_3 \) significantly increased the emergence percentage at initial stages as compared to other priming treatments. At 8\(^{th}\) DAS \( \text{GA}_3 \) induced maximum emergence of seeds. At 10\(^{th}\) day maximum emergence was recorded under \( \text{KNO}_3 \) followed by sodium selenite. On the other hand, seedling emergence started from 10\(^{th}\) day in control treatment. The final count of emergence in control treatment was about one third of \( \text{GA}_3 \) and warm water treatment. Though the emergence of seedlings of seeds treated with warm water started at 8\(^{th}\) DAS, it was
vigorously accelerated after 10th DAS (Fig.2.1) and reached to maximum as GA$_3$ and KNO$_3$ treated seeds did it.

![Graph showing the effect of different seed treatments on seedling emergence of 'uchja' seeds.]

Figure 1. Effect of different seed treatments on seedling emergence of ‘uchja’ seeds

**Seedling Vigor**

Vigor index, emergence index, and co-efficient of emergence were increased significantly in primed seeds compared to untreated control (Table 1). The highest vigour index (12.68), was found in GA$_3$ followed by KNO$_3$ (11.10), warm water (10.42), sodium selenite (9.66) and warm water (8.42) treated seeds and it was lowest in control (6.55). Emergence index were also significantly increased with priming and it was highest in GA$_3$ (33.60) followed by KNO$_3$ (31.32) and the lowest value (12.55) was found in untreated seeds. Co-efficient of emergences was statistically similar in all the treatment except untreated control (Table 1).
Table 1. Effect of different priming practices on seedling emergence and speed of germination on small type bitter gourd ‘uchja’

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Emergence (%)</th>
<th>Emergence Index</th>
<th>Vigour Index</th>
<th>Co-efficient of germination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (untreated)</td>
<td>40 d</td>
<td>12.14 e</td>
<td>6.55 c</td>
<td>6.15 c</td>
</tr>
<tr>
<td>Normal water</td>
<td>66 c</td>
<td>20.20 d</td>
<td>7.51ab</td>
<td>8.42 ab</td>
</tr>
<tr>
<td>Warm water (50°C)</td>
<td>96 a</td>
<td>27.00 c</td>
<td>10.42 ab</td>
<td>7.85 b</td>
</tr>
<tr>
<td>KNO₃ (1%)</td>
<td>94 a</td>
<td>31.32 ab</td>
<td>11.10 ab</td>
<td>8.30 ab</td>
</tr>
<tr>
<td>Sodium selenite(10 ppm)</td>
<td>90 b</td>
<td>28.00 bc</td>
<td>9.66 b</td>
<td>8.21 b</td>
</tr>
<tr>
<td>GA₃ (400 ppm)</td>
<td>96 a</td>
<td>33.60 a</td>
<td>12.68 a</td>
<td>8.51 a</td>
</tr>
</tbody>
</table>

Data followed by the same letter within a column do not differ significantly by Duncan’s Multiple Range Test (DMRT) at 5% level of probability.

Vigour index exhibited positive linear relationship with germination percentage and over 80% variation in germination percentage could be explained by the variation of vigour index (Fig. 2). Pill (1986) found partly similar result who suggested that speed of germination determines the success of crop establishment in the field condition.
Length of Shoot, Root and Dry Weight of Bitter Gourd (Uchja) Seedlings

The length and dry weight of shoot and root of bitter gourd seedlings was influenced significantly by different priming practices (Table 2). The shoot length was lowest in control which was statistically comparable to that of all the priming treatments. Shoot length increased from (49 to 85 cm). The seedling of untreated control treatment produced the lowest shoot length (32 cm) and it was highest in GA$_3$ treated seeds. In case of root length, the lowest value (7.12 cm) was attained at control and the highest (9.25 cm) in GA$_3$ treated seedling. However, the values of root length were statistically identical to all the treatments.

The shoot dry weight increased significantly with priming practices at all the treatments. It was lowest at untreated seeds (0.71 g), moderate at warm water (0.9 g), KNO$_3$ (1.3 g) and sodium selenite (1.75 g) and highest at GA$_3$ (2.12 g). This finding is an agreement with the results of Pandita and Nagarajan (2004). The primed seeds received the positive responses for germination consequently positively influenced on the seed vigour which resulted in higher seedling growth. This growth was mainly pronounced on the shoot growth despite of root, which might have less competition for soil nutrients as the seedlings mainly dependent on the seed reserve materials.
Table 2 Effect of different priming practices on shoot and root growth of small type bitter gourd ‘Uchja’

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Shoot length (cm)</th>
<th>Root length (cm)</th>
<th>Shoot dry matter (g)</th>
<th>Root dry matter (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>32 e</td>
<td>7.12 c</td>
<td>0.71 b</td>
<td>0.029</td>
</tr>
<tr>
<td></td>
<td>46 d</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal water</td>
<td></td>
<td>7.5 bc</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>49 d</td>
<td>0.9 ab</td>
<td>0.028</td>
<td></td>
</tr>
<tr>
<td>Warm water</td>
<td></td>
<td>7.15 c</td>
<td></td>
<td></td>
</tr>
<tr>
<td>KNO₃</td>
<td>59 c</td>
<td>1.3 ab</td>
<td>0.042</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>7.58 bc</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sodium selinite</td>
<td>67 b</td>
<td>1.75 ab</td>
<td>0.061</td>
<td></td>
</tr>
<tr>
<td></td>
<td>8.5 a</td>
<td>2.12 a</td>
<td>0.085</td>
<td></td>
</tr>
<tr>
<td>GA₃</td>
<td>85 a</td>
<td>9.25 a</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Means followed by the same letter (s) within a column did not differ significantly at 5% level by LSD

DISCUSSIONS

The present study revealed that a significant improvement was made on seedling emergence and seedling vigor due to priming practices of seeds of ‘uchja’. Percent emergence of seedlings varied widely among the primed and non primed seeds of ‘uchja’. It might be due to metabolic repair in plasma membrane of seeds during imbibitions, by increasing germination enhancing metabolites and build up osmotic adjustments (Basra et al., 2005). Similarly, Bradford (1986) reported that priming favoured the metabolic process necessary for germination. Primed seeds exhibited a rapid emergence and greater emergence uniformity. Because pre-sowing hydration might have softened the seed coat that allowed the leakage of germination inhibitors in the seed and this might have contributed to the enhancement of seed germination (Harris, 1996). The bitter gourd seeds rapidly lost its viability, priming might have enhanced the repair of cell membranes that were disrupted during ageing and repair of membranes could initiate reactivation or re-synthesis of membrane bound enzymes and enhanced germination (Rao and Sinha, 1993; Chlu et al., 1995).

Priming treatments increased seedling emergence by 96 % both in GA₃ and warm water, while 94 and 90 % in KNO₃ and in sodium selenite respectively as compared to 40 % at control under sub-optimum temperature(<20°C). The result clearly revealed that priming practices counteract the sub-optimum temperature for improving germination. Lin and Sung (2001) also stated that priming counteracted the low temperature effect by increasing the activity of different enzymes like isocitrate lyase, malate synthase and malate dehydrogenase involved in lipid and sucrose conversion. On the other hand, priming treatments improved both percent emergence and seedling vigor which might be partly
contributable to decreased lipid peroxidation during imbibitions (Hsu et al., 2003). Similarly, Wang et al. (2003) reported that suboptimum temperature namely below 20°C decreased germination while increased the production of malondialdehyde and total peroxides accumulation and reduced the anti-oxidative defense system in bitter gourd.

Vigor index determines the state of the health of seedling and ultimately the state of the productivity of the plant. Higher the vigor index better will be the yield of the plant. It was observed from the study that priming treatments induced significant changes in vigor index.

GA$_3$ and KNO$_3$ exhibited highest vigor index by 12.68 and 11.10. It is assumed that GA$_3$ might have increased the $\alpha$-amylase activity for breakdown of starch stored in seeds. Similarly, KNO$_3$ also might have increased the $\alpha$-amylase activity and total sugar content in seeds (Basra et al., 2005)

Treatment of seeds of ‘uchja’ with sodium selenite also improved emergence of seedlings and speed of seed germination at sub-optimum temperature. It might be linked to the enhanced activity of glutathione peroxidase (free radical and peroxide scavenging enzyme). This enzyme activity is induced by sodium selenite (Chen and Sung, 2001).

CONCLUSIONS

Seed priming treatments significantly increased seedling emergence and speed of germination and uniform germination at sub-optimum temperature. Among the different priming treatments GA$_3$ showed superior to all in respect of emergence and vigor of seedling followed by warm water treatment. However, pre-sowing seed treatment with warm water might be better for an easy and cheaper technique.

REFERENCES


