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Effects of hydropriming on seed germination, seedling growth and yield of bitter gourd

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ABSTRACT

The experiment was conducted to find out the effects of hydropriming on seed germination, seedling growth and yield of bitter. Two single factor experiments were conduced at the laboratory and field of Seed Science and Technology Department following completely randomized design with three replications and randomized complete block design with five replications, rspectively. The experiments comprised six soaking treatments viz. T_0: untreated (control), T_1: tap water (around 25° ± 2° C) for overnight, T_2:cold water (12ºC) for 5 min, T_3: cold water (12ºC) for 10min, T_4:cold water (12ºC) for 15min, T_5: hot water (45ºC) for 5 min and T_6: hot water (45ºC) for 10min soaking. The highest germination (88.0%) was obtained from T_5 treatment and the lowest germination (30.0%) was recorded in control treatment. It was also observed that T_5 treatment exehibited superior performances in respect of most of the parameters studied in the laboratory as well as in fiield experiment as compeared to control treatment. In respect of fruit yield per plant, maximum number of fruits obtained from T_5 treatment (16 fruits/plant) followed by T_6 (14 fruit/plant), T_1 (12 fruit/plant), T_2 (12.0 fruit/plant), T_3 (11.2 fruit/plant) and T_4 (11.2 fruit/plant) and the minimum number of fruits per plant recorded from the control treatment (7.6). From the findings of these study it can be concluded that bitter gourd seeds soaked in hot water at 45ºC for 5 min could improve seed germination, expedite seedling growth and eventually increase the yield of fruit.

Introduction

Bitter gourd *(Momordica charantia* L.) belongs to the family Cucurbitaceae commonly known as *Karola* in Bangladesh. It is popular due to its medicinal properties which is usually recommended for curing blood diseases, rheumatism, diabetes and asthma. According to Yibchokanun *et al*. (2006), bitter gourd is a blood purifier, activates spleen and liver and is highly beneficial in diabetes. Bitter gourd is very much helpful in curing the diabetes having anti carcinogenic properties and can be used as a cytotoxic agent against many types of cancer (Grover and Yadav, 2004). Each 100 g of edible portion of bitter gourd fruit contains 2.1g protein, 1.0g fat, 1.4 g minerals, 1.7 g fiber, 10.6 g carbohydrate, 23 g calcium, 38 g phosphorus, 2.0 g iron, 126 µg carotene, 0.07 mg thiamine, 0.06 mg riboflavin and 96 mg vitamin (Gopalan *et al*., 1982). Nevertheless, its fruits are rich in phytonutrients like dietary fibre, minerals, vitamins and antioxidants. Total vegetable production in Bangladesh was about 3448000 tons in 414979.76 ha area of land. Whereas bitter gourd production was 57000 tons with an average yield of 5.5tons/ ha in 10526.32 ha of land (BBS, 2017). This figure does not indicate a good figure of bitter gourd in comparative to total vegetables yield. Germination is a critical stage in the life cycle of weeds and crop plants and often controls population dynamics, with major practical implications (Huang *et al*., 2003). Seed germination is the critical stage for species survival (Yang *et al*., 2008). So, low percentage of seed germination may be one of the major reason behind this issue.

Hydropriming of seeds is done by soaking of seeds in water either cold, hot or normal for different duration before sowing in the field. Thick seed coat enclosing embryo affect germination by imposing mechanical restriction on embryo growth. This problem of poor or slow seed germination can be solved through many techniques and one of them is seed priming (Pandita and Nagarajan, 2007). Seed priming reduces the germination time, increases germination percentage, seedling

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emergence, and increases uniformity under adverse environmental conditions. Soaking duration and water temperature may vary depending on the type of seeds. Treated seeds show earlier germination and further growth whereas untreated seeds give late germination, smaller size plant, lower number of branching, late flowering, late fruiting, etc. being vulnerable to pathogen attack. Through water treatment of seeds the pathogenic structures can be washed out. Hot water seed treatment also has the beneficial effect of priming seeds resulting in faster germination than untreated seed. Therefore, the present study was undertaken to investigate the effects of hydropriming on seed germination, seedling growth and yield of bitter gourd**.**

Materials and Methods

Two separate experiments were conducted in the Field and in the Laboratory of the Department of Seed Science and Technology, Bangladesh Agricultural University, Mymensingh during November 2016 to March 2017. The treatments used for both the experiments were T_0: untreated control, T_1: tap water (around 25ºC) soaking for overnight, T_2: soaking in cold water (12ºC) for 5 min, T_3: cold water (12ºC) for 10 min, T_4: cold water (12ºC) for 15min, T_5: hot water (45ºC) soaking for 5 min and T_6: hot water (45ºC) for 10min. The field experiment was laid out in a Randomized Complete Block Design (RCBD) with five replications having five blocks. Each block contains 7 numbers of 30 cm x 30 cm x 30 cm size pits. Data were recorded on germination percentage, vigour percentage, primary branch length, leaf area, fresh weight of shoot and root, days to first flowering, days to first fruiting, length and width of fruit, fresh weight of individual fruit and total number of fruits per plant. The seeds of local variety of bitter gourd (*Momordica charantia* L.) having good physical appearance (uniform size, shape, color, etc.) were selected. Seeds were collected from local market. The second was performed in laboratory condition followed by aforementioned six treatments and same replication. About 30 cm x 30 cm x 30 cm size plastic pots was used for seed germination. Pots were filled with two third sandy loam soil and seeds were sown for germination. Germination percentage, seedling vigour, shoot length and leaf area were calculated from this experiment. Germination is the emergence of radical and plumule. It was calculated by regular observation of the sown seeds. It was calculated as follows (Krishnasamy, 1990):

$$\% G = \frac{S_E}{S_T} \times 100$$

Where, *% G* = germination (%), S_E and S_T designate number of emerged and total seeds, respectively.

Vigour indicates the ability of seeds how fast it produces seedling. It also gives an idea of strengthness of seedlings. Vigour index is measured by counting the normal as well as abnormal seedlings. Any deviation from normal condition is called abnormal seedlings. It was marked by visual symptom and calculated as follows (Copeland, 1976):

$$\% V = \frac{S_N}{S_T} \times 100$$

Where, *% V* = vigour (%), S_N and S_T designate number of normal and total seedlings, respectively.

The collected data on different parameters were statistically analyzed to obtain the level of significance using MSTAT-C package programme. The mean differences were compared by Duncans Multiple Range Test (Gomez and Gomez, 1984).

Results and Discussion

Germination percentage

Different hydropriming treatments showed significant differences on germination percentage of bitter gourd. Though treatment no. 5 (soaking in hot water for 5 min) gave the highest seed germination, other treatments also noted higher percent germination than the control. The highest germination percentage was 87.00 in lab and 88.00 in field condition (Table 1). Seeds soaked in cold water (12 ºC) at different duration did not show significant performance over the treatment with hot water (45 ºC) soaking for 5 min. Germination percent for T_6 and T_5 was also non-significant. The result of this parameter in case of control was 26.67% in laboratory and 30.00% for field (Table 1).

Vigour percentage

Seedling vigour percentage calculated at 20 DAS was significantly different in respect of various hydropriming treatments. The highest vigour percentage was 63.20 in lab and 66.58 in field against the treatment T_5 (Table 1). Control (non soaked seeds) gave seedling vigour 25.20% in lab and 19.12% in field followed by the lowest value. Performance of T_3 and T_4 in case of vigorous seedling production was almost similar and non-significant between them. Seeds treated with both normal water and hot water at different durations showed more vigorous seedlings than untreated seeds. Moreover, vigour percentage at T_1 (60.00% and 58.00%) and T_6 (57.89% and 57.13%) was significant over the control (Table 1).

Shoot length

The length of shoot at different intervals (both in lab and field) also responded remarkably under different water soaking treatments. Shoot length at 20 DAS was recorded at the treatment of hot water soaking for 5 min and it was 14.93 cm in lab and 16.60 cm in field and that was maximum value among the treatments (Table 2). On the other hand, the minimum length was recorded at 5.57 cm in lab and 5.68 cm in field at same DAS for control.

Table 1. Germination percentage and vigour percentage under different hydropriming treatments

Treatment	Germination (%)		Vigour (%)	
	Laboratory	Field	Laboratory	Field
T_0	26.67 d	30.00 d	25.20 c	19.12 e
T_1	60.00 b	58.00 b	42.91 b	40.41 c
T_2	50.00 bc	54.00 b	31.11 c	33.67 d
T_3	43.33 c	44.00 c	29.78 c	31.24 d
T_4	43.33 c	42.00 c	29.11 c	32.67 d
T_5	87.00 a	88.00a	63.20 a	66.58 a
T_6	76.67 a	80.00a	57.89 a	57.13 b
$LSD_{0.05}$	10.88	8.80	10.79	6.52

T_0= untreated seeds, T_1= soaking in normal tap water for overnight, T_2= soaking in cold water for 5 min, T_3= soaking in cold water for 10 min, T_4= soaking in cold water for 15 min, T_5= soaking in hot water for 5 min and T_6= soaking in hot water for 10 min. Figures followed by different letters differ significantly at 1% level of probability.

Table 2. Hydropriming treatment effects on shoot length at 20 DAS both laboratory and field experiment

Treatments	Shoot length (cm)	Shoot length (cm)
	Laboratory	Field
T_0	5.57d	5.680d
T_1	13.30b	13.06b
T_2	9.23c	10.18c
T_3	8.80c	9.780c
T_4	8.33c	8.620c
T_5	14.93a	16.60a
T_6	14.50a	15.70a
$LSD_{0.05}$	1.09	1.50

T_0= untreated seeds, T_1= soaking in normal tap water for overnight, T_2= soaking in cold water for 5 min, T_3= soaking in cold water for 10 min, T_4= soaking in cold water for 15 min, T_5= soaking in hot water for 5 min and T_6= soaking in hot water for 10 min. Figures followed by different letters differ significantly at 1% level of probability.

Table 3. Hydropriming treatment effects on primary branch length of bitter gourd at different days after sowing at field condition

Treatments	Primary branch length (cm)			
	30 DAS	45 DAS	60 DAS	70 DAS
T_0	8.240f	19.30f	31.40d	36.40e
T_1	14.38c	32.84c	46.40c	48.26c
T_2	13.90cd	28.40d	42.80c	44.80d
T_3	12.50de	22.80e	43.20c	44.40d
T_4	11.20e	21.24ef	43.40c	43.80d
T_5	22.42a	45.40a	58.60a	66.20a
T_6	20.80b	42.80b	54.20b	53.60b
$LSD_{0.05}$	1.43	2.17	3.86	2.86

T_0= untreated seeds, T_1= soaking in normal tap water for overnight, T_2= soaking in cold water for 5 min, T_3= soaking in cold water for 10 min, T_4= soaking in cold water for 15 min, T_5= soaking in hot water for 5 min and T_6= soaking in hot water for 10 min. Figures followed by different letters differ significantly at 1% level of probability.

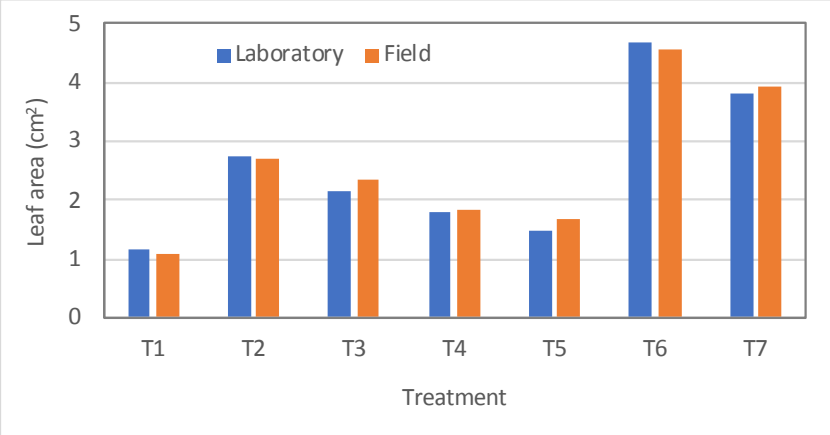

Fig 1. Hydropriming treatment effects on leaf area of bitter gourd at 20 DAS observed in laboratory (A) and field (B) conditions. Vertical bars represent LSD at 5% level of probability.

Table 4. Hydropriming treatment effects on shoot fresh weight and root fresh weight per plant at laboratory experiment

Treatments	Fresh weight of shoot (g/plant)	Fresh weight of root (g/plant)
T_0	0.378 e	0.338 b
T_1	0.753 b	0.456 a
T_2	0.582 c	0.338 b
T_3	0.529 cd	0.456 a
T_4	0.428 de	0.419 a
T_5	0.985 a	0.479 a
T_6	0.882 ab	0.472 a
$LSD_{0.05}$	0.136	0.062

Table 5. Effects of hydropriming treatments on days to first flowering and days to first fruit set of bitter gourd in field condition

Treatments	Days to first flowering	Days to first fruit set
T_0	54.80a	63.40a
T_1	46.60b	57.80bc
T_2	46.80b	60.60ab
T_3	47.60b	60.40ab
T_4	47.40b	60.60ab
T_5	44.40c	55.80c
T_6	45.80bc	56.40c
$LSD_{0.05}$	2.08	3.07

Table 6. Hydropriming treatment effects on fruit length, fruit width, fruit weight and number of total fruits per plant of bitter gourd observed in field condition

Treatments	Fruit length (cm/fruit)	Fruit width (cm/fruit)	Fruit weight (g/fruit)	No. of fruits/ plant
T_0	4.00d	2.48d	6.252e	7.600d
T_1	5.52b	3.08b	10.75c	12.00c
T_2	4.54c	2.74c	10.59c	12.00c
T_3	4.54c	2.70c	8.662d	11.20c
T_4	4.44c	2.56cd	8.560d	11.20c
T_5	7.32a	3.60a	19.67a	16.00b
T_6	5.74b	3.46a	12.84b	14.00d
$LSD_{0.05}$	0.276	0.186	0.914	7.60

T_0= untreated seeds, T_1= soaking in normal tap water for overnight, T_2= soaking in cold water for 5 min, T_3= soaking in cold water for 10 min, T_4= soaking in cold water for 15 min, T_5= soaking in hot water for 5 min and T_6= soaking in hot water for 10 min. Figures followed by different letters differ significantly at 1% level of probability.

Primary branch length

Only in field condition, length of primary branch at different days of interval was taken. The results showed significant differences among the treatments. The highest value of primary branch length was found 22.42 cm, 45.40 cm, 58.60 cm and 66.20 cm at 30 DAS, 45 DAS, 60 DAS and 70 DAS respectively in respect of treatment T_5. On the contrary, lowest values at the same DAS were 8.24 cm, 19.30 cm, 31.40 cm and 36.40 cm respectively in treatment T_0 (Table 3).

Leaf area

There was a significant variation among the hydropriming treatments observed in leaf area of bitter gourd. It was found that he highest value of leaf area at 20 DAS was 4.66 cm^2 (in field) and 4.56 cm^2 (in lab) and the lowest value was found as 1.16 cm^2 (in field) and 1.06cm^2 in laboratory (Figure 1). The results obtained for T_3 (1.83 in lab and 1.78 in field) and T_4 (1.67 in lab and 1.50 in field) was non-significant (Figure 1).

Fresh shoot and fresh root weight/plant

Fresh shoot weight /plant was taken at 20 DAS under laboratory condition. The shoot fresh weight was maximum (0.985 g) in treatment T_5 (seed soaking in hot water for 5 min) and minimum (0.378 g) in control (Table 4). The results also showed significant differences in different soaking treatments over non-soaked. Maximum root fresh weight was observed at 20 DAS was 0.479g and minimum root fresh weight was 0.338g, respectively in the treatment T_5 and T_0 (Table 4).

Days to first flowering

Variations in respect of days to first flowering were found to be highly significant for different hydropriming treatments. Days required for first flowering was recorded with frequent observation. The minimum days required for first flowering was 44.40 from days of sowing and maximum days required 54.80 days (Table 5).

Days to first fruit set

In terms of first fruit set days the variations were also found significant for different soaking temperature and durations. Days to first fruit set in respect of different treatments was recorded. The minimum days for first fruit set was 55.80 from the days of sowing and maximum days to first flowering needed for treatment T_0 was 63.40 (Table 5).

Fruit length

The difference in respect of individual fruit length of bitter gourd for the seed soaking treatments exhibited statistically significant result. Maximum length of collected mature single fruit was obtained to be 7.32 cm and the minimum length per fruit was 4.0 cm (Table 6).

Fruit width

Fruit width at the middle portion of the fruit was taken for individual fruit in respect of different treatments. Obtained maximum fruit width was 3.60 cm and the minimum were 2.48 cm in treatment T_5 and control T_0 respectively (Table 6).

Fruit weight

Weight of each representative fruit from the plant of different treatments was recorded. The highest value was obtained to be 19.67g and the lowest value was 6.25g in treatment T_5 and control T_0 respectively (Table 6).

Number of total fruits /plant

Individual fruit weight of bitter gourd significantly varied due to different treatments with different water temperature and duration of soaking. Fruits were collected for a certain period of time in three installments and the number of fruits in each installment were recorded. The collected highest number of fruits were 16.00 and the lowest value was 7.60 up to 70 DAS (Table 6). There were non-significant variations between the treatments T_1 and T_2. T_3 (11.20) and T_4 (11.20) had also produced same number of fruits per plant (Table 6). Number of total fruits per plant for the treatment T_6 (14.00) was also non-significant.

Discussion

Germination is an important prerequisite for continuation of further growth and other physiological process including production or yield. The performed experiment with six treatments showed that the treated seeds resulted better germination percentage than untreated. The best result in case of germination was found for the treatment soaking seeds in warm water (45ºC) for 5 minutes both for laboratory and field conditions. Each crop cultivar requires a critical soaking duration and it should be less than the safe limit (Harris *et al*., 2000). Having thick seed coat sometimes proper absorption of water becomes difficult in case of bitter gourd. This results in late germination and also dormancy of seed. Soaking of seeds before sowing may be beneficial in this aspect. Soaking helps to absorb water easily and also remove some surface pathogen of seeds. Seed priming, the controlled hydration and dehydration of seeds, is used extensively to increase the rate and uniformity of seedling establishment of commercial vegetable and flower seeds (Bradford, 1986; Khan, 1992).

Sometimes it is still contradictory in identifying the best treatment according to seed priming by seed soaking materials, temperature and durations for highest

germination and further growth, because both germination and growth not only depended on types of seeds but also greatly influenced by many internal and external factors. The results of the present study showed that hot water treatment of seeds before sowing gives better germination percent than normal water soaking. In winter season, bitter gourd germination becomes late due to low temperature of soil than required temperature. Hot water treatment of seeds thus allow better and earlier germination even in winter season. A range of crop species showed faster germination, early emergence and vigorous seedlings achieved by soaking seeds in water for same time followed by surface drying before sowing, which may result in higher crop yield (Harris *et al.*, 2000).

Seedling vigour means how fast a seedling can grow avoiding all adverse condition. It is count by normal seedling in comparison to total seedling. Good quality seeds give vigorous seedling. Sometimes it happens that, a seed plot may give better germination but normal seedling are very few in number. Seeds treated with both normal water and hot water at different duration showed more vigorous seedlings than untreated seeds. Some seed borne pathogens do not attack at initial stage but when seedling stage starts they grow with the growth of plant and even stop the growth in severe. Seeds with hot water treatment as well as treatment with other chemicals like potassium nitrate, gibberellic acid, methyl jasmonate, etc. with different concentrations can kill pathogen and improve seed germination. As one of the treatments gave most vigorous seedling, obviously it gave better result of shoot length, and good performance of other growth parameter like leaf area, root length etc.

Leaf blade is an important parameter in growth measurement as well as yield. It is the most active part of photosynthesis. The treated seeds when produced more healthy and vigorous seedling, the seedling or plant produced the best quality leaf either in length, breadth, color, size or shape. It may be because of earlier germination and less susceptible to pathogenic attack since treated seeds were washed with warm water. The influence of pre-sowing treatments using hydropriming, osmopriming and halopriming in laboratory and/or nursery tests on seedling emergence, seedling weight and plant growth of endive and chicory are satisfactory (Ashraf and Falood, 2005). Priming stimulates series of biochemical change in the seed that are essential to initiate the emergence process like break down of dormancy, hydrolysis, metabolism of growth inhibitors, imbibitions, activation of enzymes (Ajouri *et al.*, 2004; Farooq *et al.*, 2009; Pukacka and Rajajczak, 2005). Soaking stimulates and produces enzymes like amylase and lipase which activate storage materials in seeds and mobilize anti-oxidant enzymes which subordinate per oxidation in seeds that retain seed vigour causing earlier emergence. Rehydration causes early emergence by influencing pre-germinative processes for germination (Farooq *et al.*, 2009). Pre-sowing treatments in raya seeds encourage growth of crop, reduced days to emergence, number of branches /plant and plant yield (Ullah *et al.*, 2012).

In this study, treated seeds gave healthy vigorous seedling which ultimately ensured more number of branches than that of untreated seed. Number of branching is also important because of being cucurbits. Vine plants give flower mostly in secondary and tertiary branches. So, the plant having more branches will give more flower and fruit and will increase the yield. The primary condition of fruit is flower, because the ovary of flower will turn into fruit. Days to first flowering is important, because if a plant produce flower in less time even a single day before it will get more time for earlier pollination. The flower which bloom earlier get more chance of pollination. The earlier the pollination, the earlier the fertilization and earlier fruit set will occur. Treatment of seeds before soaking gives a positive result in case of first flowering days in bitter gourd. It has been shown that treatment of seeds with warm water gives earlier flowering than untreated seeds. Seed soaking of tomatoes minimizes days of flowering (Murungu *et al.*, 2004). Rice priming causes earlier flowering in the crops and enhanced the yield (Harris *et al.*, 2000). Priming also enhances the activities of anti-oxidative enzymes in treated seeds (McDonald, 1999; Wang *et al*., 2003). Moreover, priming increases the activities of glyoxysome enzymes in primed bitter gourd seeds (Mehta, 2014).

Hydropriming is a very simple, economic and environmental friendly type of seed priming (Jamil *et al.*, 2016). Not only the number of fruit is important but also the weight, size, etc. of fruit are considering point in case of good production of a crop. Every living organism wants to keep its progeny or identity. The plant having poor growth may also give a good number of fruits. But if the mother plant is not healthy it can't produce healthy offsprings. So, these fruit obviously be deformed in shape and small in size. From different experiments it has been observed that vigorous plant from treated seed gave larger and uniform shaped fruit. In this study the best result was found in case of seed soaking at 45ºC for five minutes. Plants from treated seeds became healthier and gave earlier flower as well as more fruit per plant than plants from untreated seeds. Plant from untreated seeds are more susceptible to seed borne pathogens, whereas plant from treated seeds can avoid this attack by initial killing or inactiveness of pathogenic structure before sowing through treatments. After all, the plants produced from untreated seeds gave poorer result than the plants of any treated seeds, either normal water, cool water or hot water.

Conclusion

In conclusion, hydropriming of bitter gourd seeds has the potential to enhance seed germination, seedling growth and also yield of bitter gourd. Seed soaking in hot water (45ºC) for 5 min effectively enhanced germination percentage, seedling vigour, other growth parameters

such as primary branch length, leaf area, etc. and the reproductive characters like days to first flowering, first fruit setting and total number of fruits per plant, while the other water soaking treatments at different temperatures and duration also exhibited better performance than the untreated control seeds. The results of this study indicated that hydropriming is a useful for improving seed germination and higher yield of bitter gourd.

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