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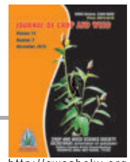

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Effects of various concentrations of boron on germination, seedling growth and sugar mobilization in wheat using boric acid primed seeds

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ABSTRACT

In the present piece of experiment wheat seeds of variety HUW -468, primed with four different concentrations (ranging from 2.0 to 10 mM) of Boric acid (H_3BO_3) used as the source of boron (B) to find out its effect on germination. Various parameters like germination percentage, final germination percentage, mean germination time, germination index, germination rate index, coefficient of velocity of germination, mean germination rate, plumule, radicle and seedling lengths, fresh and dry weights of radicle and plumule, α-amylase activity and soluble sugar content of/in endosperm/seed respectively were determined at 3,5,7 and 9^{th} days during germination and seedling growth. Boric acid primed seeds showed better performance for all the parameters in respect to non- primed seeds. Among treatments 8 mM (T_3) has presented the best result than the other three [2(T_1), 4(T_2) and 10(T_4) mM] used concentrations of boric acid and non-primed control (T_0) sets. α-amylase activity was increased steadily up to 7^{th}day and then declined on 9^{th} day in which T_3 showed the highest activity of enzyme as regard to other four treatments. Soluble sugar content was lower on 3^{rd} day, but it started to increase from 5^{th} day onwards and again declined on 9^{th} day in endosperm of germinating seeds. Report suggested that 8 mM concentration of boric acid equivalent to 86.48 mg l^{-1} of boron in boric acid solution can improve the germination process of wheat variety via facilitating the solubilization and mobilization of sugar in endosperm which may be due to the presence of micronutrient boron.

Keywords: α-amylase activity, micro-nutrient, nutripriming, boron, seedling vigour, soluble sugar content

A technique known as seed priming accelerates the uniform and quick emergence of seeds during germination in order to produce seedlings with high vigour and faster crop growth. The technology's operation is dependent on the ions, chemicals, and plant growth regulators utilised, all of which are present in the medium of the seed priming system. It is generally known that seed priming enhances germination physiology and equips seeds to withstand a variety of stressors (Bose *et al.,*2018). Even when wheat is exposed to high metal stress, seed priming generally seems to boost germination responses (Kumar *et al.,* 2016). According to Anaytullah and Bose (2007) , seed priming with Mg $(NO_3)_2$ and KNO_3 can alleviate the effects of low temperature stress. To prepare seeds for stressful conditions during germination and plant growth, seed priming appears to trigger an osmotic adjustment during the process of hydration/dehydration within the seed (Bradford, 1986). According to Bose and Mondal (2013), chemicals employed in nutripriming, such as NaCl, KH_2PO_4, K_3PO_4 and KCl have a modifying effect on plant metabolic processes. According to Jangde *et al.* (2014), wheat seeds treated with Ca^{2+} salts such $CaCl_2$ or $Ca(NO_3)_2$ during the cold temperature germination period showed improvements in germination percentage, relative and absolute water content, and α-amylase activity. According to research by Murata *et al.* (2008), calcium salt seed priming can improve maize seedling emergence while lowering seedling mortality. Mondal and Bose (2019) claimed that micronutrient seed priming improves the germination rate, seedling establishment and various physico-chemical and molecular processes during the growth and development of crops, but the extent of its effect depends on the type of nutrients used as well as their concentration.

It is generally known how crucial certain micro-nutrients are to a plant's survival. As a micronutrient, boron (B) is involved in numerous metabolic processes that occur throughout the growth and development of crops. A crucial micronutrient for plant growth and development, iron plays a role in the movement of sugar, the germination of pollen grains, and plant reproduction. For crops to grow in both quality and quantity, as well as during flowering and fruiting, boron is necessary. Numerous metabolic activities, including the production of cell walls and the maintenance of membrane integrity, involve the mineral borax (Tanaka and Fujiwara, 2008). Reddy *et al.* (2007) observed that seed priming with

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boron was more successful than control at increasing seed yield in pigeon pea.

The time to germinate, ultimate germination percentage, germination energy, germination index and mean germination time in the rice crop were all increased by seed priming with B solutions (Farooq *et al.*, 2011). According to Rehman *et al*. (2012), the use of boron as a seed priming agent improves seedling emergence, leaf appearance and elongation, chlorophyll content, water relations and yield-related traits of fine rice, but it is strictly concentration dependent. For example, B at a concentration of 0.5% failed to germinate the rice seeds, whereas 0.001 to.0.1% improved seedling stand (Rehman *et al.,* 2012a). The development, emergence and yield of wheat seedlings are typically improved by boron seed priming (Iqbal *et al.,* 2017). Numerous reports claimed that applying boron externally enhanced numerous crops' germination, growth, yield, quality and nutrient content. However, there are few findings on how boron treatment within the seeds of different wheat varieties during germination affects modifications or increases in metabolic events. Therefore, the goal of the current study is to determine the best concentration of boron, in the form of boric acid, to use during the germination of the wheat variety HUW-468 and to determine whether boron application during seed germination can enhance the ability of seed to mobilise and solubilize carbohydrates.

MATERIALS AND METHODS

The Department of Plant Physiology's seed priming research laboratory at the Institute of Agricultural Sciences, Varanasi served as the site of the current inquiry. The Department of Agronomy at the same institute provided the wheat seeds for the HUW-468 variety that were employed in the current experiment. The seeds were sterilised using 0.1% $HgCl_2$ solution for 3 minutes, thoroughly washed with distilled water for 4-5 times, and then let to soak for 12 hours in a variety of boric acid concentrations in laboratory at 22±2°C. Seeds that had been soaked in boric acid were dried naturally by maintaining them in an environment with forced air and a fan until they regained the weight they had before the soaking. Studies on germination and seedling development were conducted in a lab setting using Petri plates up to 9th day. The treatments were T_0 = 0 mM, T_1 = 2 mM, T_2 = 4 mM, T_3 = 8 mM, and T_4 = 10 mM of boric acid, which correspond to 0, 21. 62, 43. 24, 86. 48. and 108.1 mgL^{-1} of boron in boric acid solutions, respectively, where T_0 represents non primed seeds.

Seed germination and measurement of growth

To start this experiment 100 seeds were taken in Petri-plates and four replications were made for each 5 treatment. The petri-plates were kept at 22±2°C in the laboratory to take observations regarding seed germination at 24, 48, 72, 96 and 120 hours and germination percentage was drawn on the basis of conventional method using the following formula :

$$\text{Germination \%} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds sown}} \times 100$$

Various germination parameters were also calculated using the following formulae respectively:

i) $\text{Final Germination Percentage (FGP in \%)} = \frac{\text{Total no. of seeds germinated on end day}}{\text{no. of seeds present in petri-dish}} \times 100$

ii) Mean Time Germination (MTG in days) = $\frac{\Sigma F * x}{\Sigma F}$

Where F is the number of seeds germinated on day x and the formula was given by Al-Mudaris (1998)

iii) Germination Index (GI) = GI = (3×n1) + (2×n2) + · · · + (1 × n4),

n1= one day germinated seed, n2= second day germinated seed, n3= third day germinated seed, n4= fourth day germinated seed; and formula given by Ranal *et al*. (2009)

iv) Germination Rate Index (GRI%/day) = G1/1 + G2 / 2 +· · ·+ Gx / x,

G1 = Germination percentage ×100 on the first day after sowing, G2 = Germination percentage ×100 on the second day after sowing and the formula was given by Al-Mudaris (1998)

v) Coefficient of velocity of germination (CVG)=(N1 + N2 + — +Nx/N1T1 + -- + NxTx) × 100, N = no. of seeds germinated each day, T=no. of days from seeding corresponding to N and the formula was given by Al-Mudaris (1998)

vi) Mean germination rate (MGR)=CVG/100 and it was calculated by the formula given by Ranal *et al*. (2009)

The seeds were supposed to be germinated when the radicle has come out from the seed coat.

The length of the seedling was calculated using a centimeter scale. Studies were conducted using 4 replications of each treatment to measure the fresh weight of the radicle and plumule at 3,5,7 and 9 days. For each treatment, weight of 5 samples were taken, and their average weight was calculated by dividing it with 5. Using the traditional procedure, all of the growth studies were conducted on the same days. However, samples of radicle and plumule were held for an hour in an oven preheated to 100–110°C for killing purposes in order to determine dry weight. Then, in order to maintain a steady weight, it was placed in an oven set at 60±2°C.

To determine Seedling vigour I and II, the following formulas were taken into consideration:

Seedling vigour I : Germination%× (shoot length + root length)

Seedling vigour II : Germination% × seedling dry weight

To verify the impact of boron, utilised as boric acid, on the germination of wheat seeds, two biochemical indices, namely α-amylase and soluble sugar, were identified. The Bernfeld (1955) method was used to assess the enzyme-amylase's (EC3.2.1.1) in vitro activity in wheat endosperm. Both biochemical parameters were examined at the 3,5,7 and 9th days to determine the amount of soluble sugar present in the endosperm using the method developed by Dubois *et al.* (1956). Statistical analysis was carried out in accordance to the experiment's specifications.

Statistical analysis

Four independent replications of each treatment were used to calculate the means, and the statistical package for social science (SPSS Version 16.0) was utilised to do a variance analysis. Using the Tukey HSD test, significant differences between several treatments were identified.

RESULTS AND DISCUSSION

Table 1 represented the germination percentage of wheat seed variety HUW-468, primed with various concentrations of boric acid. The result showed that primed seeds performed well than the controlled one; at 24 and 48 hours T_3 showed better germination rate i.e., 93 and 95.75% respectively than others, followed by T_2, T_1, T_4 and T_0. At72 and 96 hours during germination T_4 showed the better germination percentage than T_1 which was followed by T_0 whereas T_3 showed better performance among all. At 120 hours of germination, T_3 showed 98.50% germination, represented the best value followed by T_1, T_4, T_2 and T_0 respectively and T_0seemed to have poor performance. Therefore in the present case 8 mM concentration of boric acid was found to be the best among used concentrations (Table 1).

Table 2 presented various germination parameters like Final germination percentage (FGP), Mean germination time (MGT), Germination index (GI), Germination rate index (GRI), Coefficient velocity of germination (CVG) and Mean germination rate (MGR). T_3 has maximum value for FGP (98.5) whereas T_0 has the least value (92.25) for FGP. MGT represents the speed of germination in which T_0has maximum value (1.257453) while T_2 has the least value (1.070681), therefore T_0 has lowest speed of germination whereas, T_2 has highest speed of germination. GI represents both germination percentage as well as germination speed and in this parameter T_3 showed maximum value (480.5) and T_0 has the lowest value (437.5), it is observed that when both the parameters are combined together like germination percentage and germination speed, T_3 has shown the best result. GRI represents the percentage of germination on each period of germination studies. The maximum (95.0375) and minimum (84.59167) values of GRI were represented by T_3 and T_0 respectively. CVG indicates the rapidity of germination where T_2 and T_0 noticed to have the maximum (93.39) and minimum (79.52) values of CVG respectively. MGR represents the rate of germination in which T_2 has the maximum value of 0.93 while T_3 and T_4 has the same values of 0.89 and T_0 has the least value of 0.79. Among all the observed germination parameters, T_3 and T_2 has shown the best result in FGP, GI, GRI and CVG, MGR respectively, whereas T_0 has shown the maximum value of MGT which indicates the slowest speed of germination among all the parameters. Thus, the result showed primed sets performed better than the non-primed one.

Boron is an important micronutrient, having multifarious activities in modulating the growth processes in plants starting from germination to yield, listed by Mondal and Bose (2019) recently. Therefore it might be also true if anyhow the boron concentration becomes optimum or even more than threshold level may influence the germination and related processes as per its nature of action.

In the present study, Table 1and 2 showed that boric acid primed sets are better performer than the non-primed control. Among the used concentrations of boric acid, 8mM concentration was found to have maximum values as regards to various germination parameters in respect to the other used treatments. The work is in accordance with the studies of Iqbal *et al.* (2012) who have noted that 0.01 and 0.001% B solutions have the ability to improve the germination of wheat cultivar

Table 1: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on germination percentage at different hours as investigated

	Germination percentage				
Treatments	**24 hrs**	**48 hrs**	**72 hrs**	**96 hrs**	**120 hrs**
T_0	80.00±0.41b	84.75±0.48b	90.75±0.75c	90.75±0.75c	92.25±0.25c
T_1	91.50±0.50a	92.50±1.85a	94.00±0.41b	94.00±0.41b	96.75±0.25b
T_2	92.00±0.71a	93.50±1.19a	95.25±0.48ab	95.25±0.48ab	95.50±0.96bc
T_3	93.00±0.71a	95.75±1.31a	96.75±0.25ab	96.75±0.25ab	98.50±0.29ab
T_4	91.25±0.48a	92.00±0.82a	95.00±0.41a	95.00±0.41a	96.00±0.41a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T_0, T_1, T_2, T_3, T_4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.

Table 2: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on various germination parameters at different hours

Treatments	**FGP (%)**	**MGT (Days)**	**GI**	**GRI (%/day)**	**CVG**	**MGR**
T_0	92.25	1.26	437.50	84.59	79.53	0.79
T_1	96.75	1.16	468.25	93.01	86.19	0.86
T_2	95.50	1.07	470.75	93.32	93.39	0.93
T_3	98.50	1.12	480.50	95.04	89.14	0.89
T_4	96.00	1.11	468.75	92.78	89.51	0.89

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T_0, T_1, T_2, T_3, T_4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.
FGP= Final Germination Percentage, MGT= Mean Germination Time, GI= Germination Index, GRI= Germination Rate Index,CVG= Coefficient Velocity of Germination, MGR= Mean Germination Rate

Mairaj-2008 and Faisalabad-2008. Induction in germination percentage was studied by Bam *et al.* (2006), who proposed that seed priming triggered the activity of hydrolytic enzymes inside the embryo and hence, hastened the germination metabolism. Seed priming enhances the process of germination that leads to increases in the rate of germination (Dahal *et al.* 1990). Matthews and Khajeh (2006) used MGT to identify the germination time in maize seedling grown in field**.** In our experiment it was observed that non primed set has more MGT than primed sets, which indicates that primed sets have germinated earlier and faster than the non-primed sets.

The data regarding lengths of plumule and radicle of germinating seedlings were presented in Table 3. In 3rd day of study it was observed that T_1 had more plumule length (0.58 cm) than T_2 (0.53 cm) but on the 5th, 7th and 9th day T_2 showed more plumule length, i.e., 2.65, 6.03 and 8.50 cm respectively followed by T_1 i.e., 2.58, 5.95, 8.35 cm. However, among all the five treatments, T_3 showed maximum plumule length at all the four intervals but T_0 presented the least plumule length. In case of radicle, T_2 has presented maximum length thanT_1 at 3, 7 and 9th day of study, followed by T_4 and T_0. At 5th day T_1 and T_2 had the same radicle length i.e., 2.95 cm followed by T_4 and T_0.However, among all the five treatments T_3 showed more radicle length at 3, 5,7 and 9th day, depicting the best boron concentration i.e., 8mM of boric acid.

By combining these two factors, plumule length and radicle length, the seedling length was finally calculated in which T_3 showed the better result than T_2 which was followed by T_1 and T_4, while T_0 showed the poorest data in respect of both radicle and plumule lengths at 3, 5, 7 and 9th day of studies, which ultimately resulted in least seedling length among all these five treatments. So by measuring the seedling length it can be said that T_3 (8mM) provided better seedling growth during all the four observed dates. Low concentration of boric acid also increased seedling length in respect to control (Table 3).

Table 3 presented plumule and radicle length, which showed that seeds primed with 8mM boric acid have the maximum values for both the parameters. The non-primed control seeds represented a poor performance in respect to primed one. Bonilla *et al.* (2004) reported that boron and calcium increased seed germination and seedling development under salt stress. Rehman *et al.*

Table 3: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on plumule, radicle, and seedling length at various stages of seed germination/seedling growth

Treatments	3rd Day			5th Day			7th Day			9th Day		
	PL	RL	SL	PL	RL	SL	PL	RL	SL	PL	RL	SL
T_0	0.33±0.02d	0.13±0.02c	0.45±0.03d	2.20±0.17b	2.28±0.06c	4.48±0.06c	4.75±0.32b	5.43±0.14b	10.18± 0.44b	7.25±0.18d	8.95± 0.15d	16.20±0.30c
T_1	0.58±0.03cd	0.25±0.02bc	0.83±0.04cd	2.58±0.14ab	2.95±0.08bc	5.53±0.05bc	5.95±0.42ab	5.90±0.18ab	11.85±0.51b	8.35±0.22cd	9.85±0.20cd	18.20±0.34c
T_2	0.53±0.03bc	0.33±0.03b	0.86±0.02bc	2.65±0.11ab	2.95±0.08b	5.60±0.04b	6.03±0.47ab	6.15±0.24ab	12.18±0.57b	8.50±0.23bc	10.05±0.16bc	18.55±0.27b
T_3	0.68±0.04ab	0.43±0.03ab	1.10±0.07b	3.03±0.12ab	3.65±0.11b	6.68±0.08b	7.03±0.49b	7.20±0.27ab	14.23±0.59ab	9.23±0.25ab	10.98±0.23b	20.20±0.33b
T_4	0.40±0.04a	0.23±0.02a	0.63±0.05a	2.33±0.19a	2.63±0.04ab	4.95±0.06a	5.83±0.38b	5.80±0.19a	11.63±0.47a	7.65±0.19a	9.28±0.18a	16.93±0.24a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T_0, T_1, T_2, T_3, T_4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.
PL= Plumule Length, RL= Radicle Length, SL= Seedling Length

Table 4: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on fresh weight of plumule and radicle at various stages of seed germination/seedling growth

Treatments	3rd Day		5th Day		7th Day		9th Day	
	FWP	FWR	FWP	FWR	FWP	FWR	FWP	FWR
T_0	13.60±0.28b	10.05±0.16c	19.85±0.27d	13.95±0.33d	25.95±0.41d	33.30±0.29d	38.20±0.52d	38.20±0.38d
T_1	16.90±0.42b	12.75±0.15b	23.95±0.17c	21.05±0.28c	33.25±0.37c	35.65±0.32cd	40.75±0.31cd	41.40±0.31c
T_2	16.75±0.37a	14.00±0.14b	24.90±0.25c	22.05±0.25b	35.30±0.38 c	36.60±0.33bc	44.10±0.41c	41.95±0.47b
T_3	18.00±0.48a	16.25±0.15b	27.10±0.12b	24.35±0.35b	37.95±0.47b	39.20±0.27b	46.90±0.43b	44.70±0.40b
T_4	13.95±0.20a	12.30±0.19a	23.55±0.18a	19.05±0.37a	32.60±0.36a	34.40±0.24a	39.60±0.53a	39.80±0.46a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T0, T1, T2, T3, T4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.
FWP= Fresh weight of plumule, FWR= Fresh weight of radicle

Table 5: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on the dry weight of plumule and radicle at various stages of seed germination/seedling growth

Treatments	3rd Day		5th Day		7th Day		9th Day	
	DWP	DWR	DWP	DWR	DWP	DWR	DWP	DWR
T_0	0.58±0.04d	0.66±0.04c	1.26±0.01d	1.00±0.02d	2.30±0.02d	2.19±0.01d	5.82±0.01e	2.98±0.05c
T_1	0.82±0.02c	1.03±0.02b	1.74±0.03c	1.64±0.03c	2.59±0.01c	2.50±0.03c	6.56±0.02d	3.45±0.03b
T_2	1.04±0.03c	1.06±0.04b	1.86±0.04b	1.65±0.01b	2.71±0.02c	2.83±0.03c	6.74±0.01c	3.46±0.03b
T_3	1.77±0.04b	1.30±0.02b	2.13±0.01b	1.93±0.03b	2.92±0.03b	3.08±0.02 b	6.89±0.03b	3.73±0.04b
T_4	0.78± 0.03a	0.93±0.03a	1.55±0.04a	1.32±0.02a	2.51±0.01a	2.45±0.01a	6.38±0.03a	3.35±0.05a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at P < 0.05. Where: T0, T1, T2, T3, T4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.

DWP= Dry weight of plumule, DWR= Dry weight of radicle

(2012b) reported that boron seed priming improves growth and yield of fine grain aromatic rice. Iqbal *et al*. (2012) reported improvement in germination as well as shoot and root length of wheat seeds primed with boron. Therefore, these statements also favor the present investigation.

The study regarding the fresh and dry weights of plumule showed that T_1 has more fresh weight of plumule than T_2 in 3rd day, whereas on 5, 7, 9th day of studies revealed that T_2 has more weight than T_1 followed by T_4 and T_0. However T_3 has represented maximum values, at all the study periods. In case of radicle fresh weight, T_2 has more values than T_1 followed by T_4 and T_0, whereas T_3 has shown the maximum value for the radicle fresh weight (Table 4).

The study of dry weight of plumule revealed that T_3 has the maximum value which was followed by T_2, T_1, T_4 and T_0 at 3, 5, 7, 9th day of studies. The same pattern was observed regarding dry weight of radicle during the same period of studies (Table 5).

Fresh and dry weights of plumule and radicle indicated same trend in the present piece of work (Table 4 and 5) which represented a better performance in case of boron primed sets as compared to non-primed one. Therefore it can be predicted that boron application in the form of seed priming has a positive impact on germination and growth of seedling of wheat variety under study. It is matched with the result of Ashagre *et al*. (2014) who observed that wheat seeds (cultivar Danda'a) primed with 0.25 mg l^{-1} of boron increased the seedling performance by increasing the fresh and dry weights of shoots and roots and more than this concentration has shown phytotoxicity. In present piece of work, 8mM of boric acid equivalent to 80 mg boron per litre is found to be the best concentration, to enhance the seedling performances of wheat variety HUW-468 when growing in petriplates under lab conditions (Table 3,4 and 5), this may resemble the importance of varietal difference.

Table 6 and 7 represent the vigour of the seedlings raised from the boric acid primed wheat seeds in terms of the index I and II. Both of the parameters presented the same pattern where T_3 concentration of boric acid, i.e., 8mM was found to be the best concentration in improving the seedling vigour and was followed by T_2, T_1, T_4 and T_0. Table 6 and 7 were indexed as seedling vigour I and II respectively. Memon *et al*. (2013) reported the highest germination index (6.289) and seedling vigour index (1753.3) in boron primed broccoli seedlings. Iqbal *et al.* (2017) also reported that boron seed priming improves seedling vigour, emergence and grain biofortification in wheat. The improvement in the germination percentage and the corresponding growth of the seedling in terms of vigour index I and II in the

Table 6: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on seedling vigour index l at various days of seed germination/seedling growth.

Treatments	Seedling vigour index l			
	3rd Day	5th Day	7th Day	9th Day
T_0	38.18±0.29d	401.33±0.43	922.98±0.52c	1494.28±0.45d
T_1	71.83±0.37cd	516.50±0.57	1113.78±0.56bc	1747.23±0.41c
T_2	84.05±0.46bc	529.20±0.33	1160.48±0.48bc	1771.55±0.58b
T_3	105.43±0.41b	644.33±0.42	1376.25±0.39b	1989.68±0.55b
T_4	57.45±0.27a	467.70±0.31	1104.50±0.44a	1637.38±0.41a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T0, T1, T2, T3, T4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.

Table 7: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on seedling vigour index II at various stages of seed germination/seedling growth

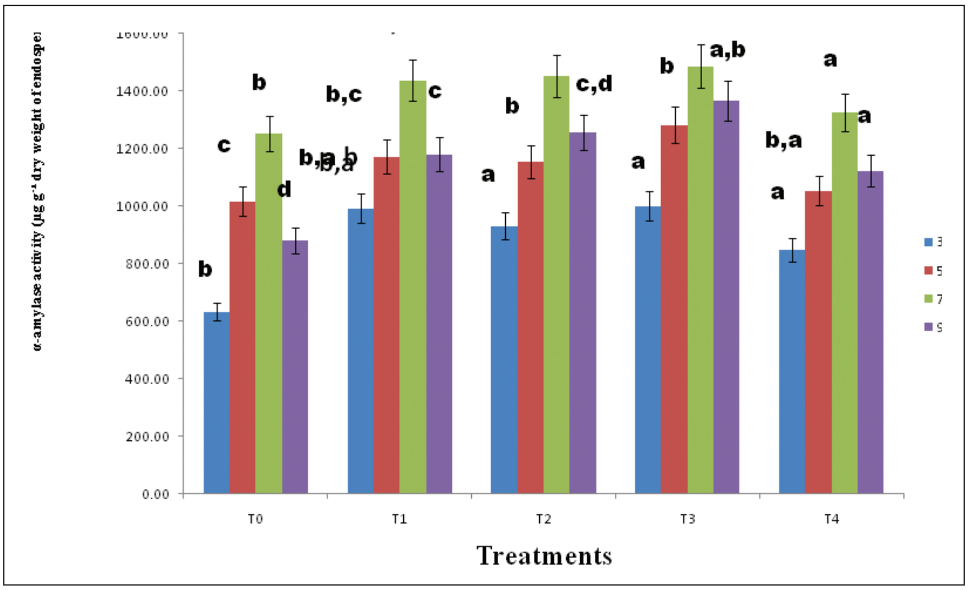
Treatments	Seedling vigour index lI			
	3rd Day	5th Day	7th Day	9th Day
T_0	104.67±0.42d	202.89±0.43d	406.97±0.56d	811.77±0.46d
T_1	170.50±0.57c	315.57±0.55c	478.48±0.48c	960.98±0.35c
T_2	196.40±0.56c	331.70±0.49b	527.65±0.39c	974.00±0.55bc
T_3	293.77±0.45b	390.81±0.43b	579.53±0.33b	1045.58±0.42b
T_4	157.19±0.51a	271.18±0.59a	471.20±0.44a	941.37±0.39a

The data are mean with standard errors from four replicates. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at $P < 0.05$. Where: T_0, T_1, T_2, T_3, T_4 are non-primed (control) seeds, seeds primed with 2mM, 4mM, 8mM and 10mM boric acid, respectively.

present case may suggest that the proper concentration of boron if employed in the form of seed priming, may also improve the mobilization of sugar from endosperm to the growing embryo as per its metabolic efforts and plays its role as an essential micronutrient for plant growth. It might be possible that during priming treatment boron may enter to seed and which is utilized during the process of germination by improving solubilization and mobilization mechanism from endosperm to growing embryo/plantlets.

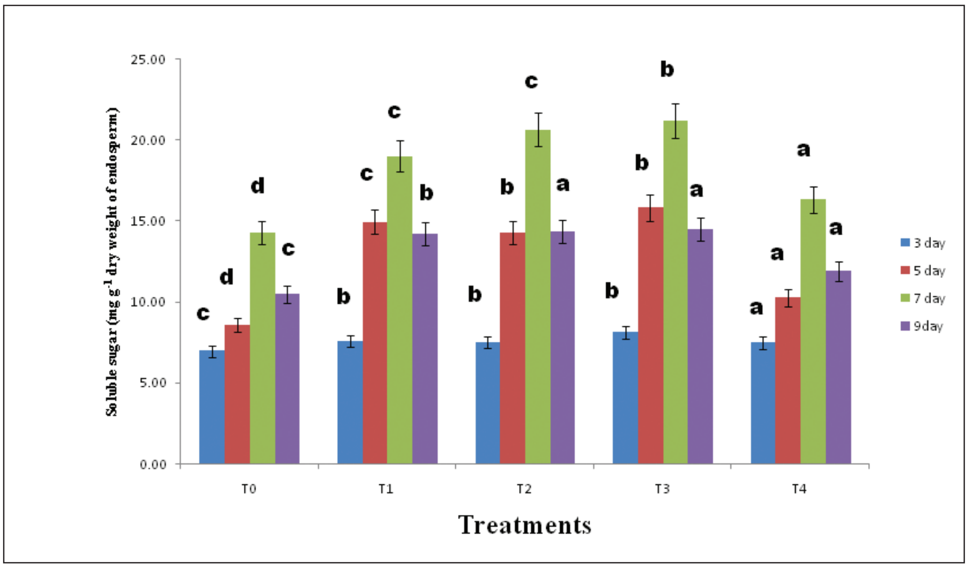
Fig. 1 represented the α-amylase activity of the enzyme extracted from 3, 5, 7 and 9th days old-germinated endosperm of wheat seeds. T_1 has shown more α- amylase activity than T_2 during the 3rd and 5th day of study which was followed by T_4 and T_0, whereas at 7th and 9th day T_2 represented more α-amylase activity than T_1 followed by T_4 and T_0. T_3 has always depicted the maximum value among all the treatments. However, the activity of hydrolyzing α-amylase enzyme increased steadily from 3rd day upto 7th day, but from 7th to 9th day it started to decline.

Study regarding the changes in the contents of soluble sugar in the endosperm of wheat seeds during germination clearly represented that treatment T_3 has the highest soluble sugar content among all the four treatments. However, on the 3rd and 5th day, T_1 has shown more soluble sugar than T_2 followed by T_4 and T_0 whereas on 7th and 9th day, the soluble sugar content in endosperm was more in T_2 than in T_1 followed by T_4 and T_0 (Fig. 2). Soluble sugar content tend to increase from 3rd day upto 7th day, but after 7th day it started to decrease upto 9th day. From the data based on α-amylase activity and soluble sugar content, it is clearly observed that both α-amylase activity and soluble sugar content started to increase from 3rd day upto 7th day but decreased 7th day onwards upto 9th day. Hence, it was noted that increase in α-amylase activity was directly proportional to increase in soluble sugar content. Fig. 1 and 2 presented α-amylase and soluble sugar content, both of them tend to increase rapidly from 3rd day of study to 5th day and attained the peak at 7th day. But after attaining a peak on 7th day it started to decline upto 9th day which entails a direct relation between α-amylase and soluble sugar content. From the study of the activity of α-amylase enzyme and the soluble sugar showed that boron may improve the activity of enzyme α-amylase may be via improving its synthesis or activity in the boric acid primed wheat seeds as a result, soluble sugar content in the germinating seeds was observed to increase with the increasing day of germination as found in the present



The mean from four replicates are shown in the figure along with standard errors. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at P 0.05. Where: T0, T1, T2, T3, T4 are non-primed (control) seeds, seeds primed with 2m, 4mM, 8mM and 10mM boric acid, respectively.

Fig. 1: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on ∞-amylase activity at various stages of seed germination



The mean from four replicates are shown in the figure along with standard errors. Different letters within each treatment denote statistically significant variations according to the Tukey HSD multiple range test at P 0.05. Where: T0, T1, T2, T3, T4 are non-primed (control) seeds, seeds primed with 2m, 4mM, 8mM and 10mM boric acid, respectively.

Fig. 2: The effect of different concentrations of boric acid used for priming wheat seeds (variety HUW-468) on soluble sugar content at various stages of seed germination.

study. Increment in α-amylase activity tend to increase the soluble sugar content in wheat var. HUW-234 and HUW- 468 with increasing days of germination in nitrate primed and non-primed control seeds (Anaytullah and Bose, 2007). Priming treatment increased α-amylase activity in the wheat kernel and establish a strong positive correlation between α-amylase and soluble sugar content. Lee and Kim (1999) showed that the plumule and radicle of primed seeds germinated and developed faster than non-primed one; they found a decrease in content of sucrose, maltose and raffinose whereas, glucose, fructose and α-amylase activity tend to increase more rapidly in primed rice seeds var. *Oryza sativa* L cv pumbyco. α-amylase activity was positively correlated with the germination rate and total sugar content (Lee and Kim, 2000). Jangde *et al*. (2014) observed an increase in α-amylase activity in wheat endosperm treated with calcium salts. Seed priming either increases the activity of α-amylase to perform better or it initiates the synthesis of new α-amylases (Sung and Chang, 1993). Therefore, the overall result suggests that boron when used as priming agent to wheat seed may improve its germination via increasing the activity/synthesis of the α-amylase enzyme, which in turn facilitates the solubilization of endosperm's storage carbohydrate. Further it induces the mobilization of sugar to the growing seedling which can be manifested with the enhanced vigor index in boron primed seeds in the present case.

CONCLUSION

In case of boron priming it might be possible that boron may enter to the seed during the process of priming and it starts to work on sugar metabolism by inducing the startup mechanism of germination. The treatment T_3 which depicts 8 mM concentration of boric acid equivalent to 86.48 mg l^{-1} of boron in boric acid solution was found to be the best concentration among the all five treatments (4 primed+1 non primed). Therefore the work concludes that the use of boric acid primed seeds of wheat may be helpful in producing vigorous seedling in the field. However, it needs a lot of future work.

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