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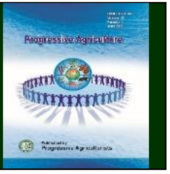

# Boron-mediated aluminium stress tolerance under aluminium toxicity at germination and early seedling stages of wheat

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## ABSTRACT

Aluminium stress is one of the major problems of wheat production that significantly reduces the growth and development. Al toxicity can be recovered by exogenous application of different growth inducing nutrient elements. Thus, this study was conducted to evaluate the ameliaration effect of B under Al toxicity. Therefore, a petri dish and hydroponic culture experiment of wheat was conducted at Crop Physiology Laboratory, Department of Crop Botany, Bangladesh Agricultural University, Mymensingh during the period from January 2016 to February 2017 to investigate the effect of boron on amelioration of aluminium toxicity in germination and seedling stage. Both the experiments were designed in completely randomized design (CRD) with three replications. The experiments were comprised of four levels of boron and aluminium concentrations *viz*., 0 µM B + 0 µM Al (control), 0 µM Al + 40 µM B, 200 µM Al + 40 µM B and 0 µM B + 200 µM Al and five wheat varieties *viz*; BARI Gom-23, BARI Gom-24, BARI Gom-28, BARI Gom-27 and BARI Gom-30. Results indicated that germination percentage, radicle and plumule length, root and shoot length, leaf length, fresh and dry mass plant$^{-1}$ were greater in 0 µM Al + 40 µM B treated plants than 0 µM B + 200 µM Al induced conditions. It indicates that wheat seedlings are susceptible to aluminium and boron can ameliorate aluminium toxicity. However, among the studied varieties, the reduction of dry mass under aluminium stress was minimum in BARI Gom-28 followed by BARI Gom-23 indicating BARI Gom-28 was more tolerant to aluminium stress than the other varieties. On the contrary, the varieties, BARI Gom-27 and BARI Gom-24 were more susceptible to aluminium stress. So it indicates that aluminium stress severely affects the growth and developments especially in the sensitive varieties and tolerant varieties have the self-ability to grow and develop even under aluminium stress condition.

## Introduction

Aluminium (Al) is the most abundant metal in the earth's crust, comprising about 8% of its mass (Tamman *et al.,* 2018). The presence of Al ions is the major constraints on crop production in acid soils, which cover more than 30% of agricultural land around the world (Agegnehu et al., 2021). On worldwide basis there are nearly 2.6 billion ha of strongly acid soils with Al$^{3+}$ toxicity (Dudal 1980; Gupta et al., 2013). Al stress

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mainly affects the soil with low pH (Gupta *et al*., 2013) and there are more than one million ha of land in Bangladesh is associated with low pH. In acid soils, Al concentration increases by increasing $Al^{3+}$ solubility at the rhizosphere zone of the plant (Iqbal et al., 2012). Hence, Al toxicity primarily affects the plant root by inhibiting root growth (Foy *et al*. 1978; Kochian *et al*., 2004) and inhibition of root elongation is the major symptom of aluminium toxicity (Ma and Furukawa, 2003; Rahman and Upadhyaya, 2021), which becomes apparent within a short period and even a small concentration of aluminium may induce root growth inhibition (Kochian *et al*., 2004). The roots are the most sensitive part of the plant (Eticha et al., 2005) and more recently, the transition zone or distal elongation zone of the root has been regarded as the most sensitive to aluminium (Li et al., 2016). Generally, root elongation is characterized by cell division and cell elongation, nevertheless, interruption of cell elongation is considered a major cause of inhibition of root growth. Aluminium toxicity also reduces the uptake of water, nutrients and interferes with cellular activities in sensitive species (Ma and Furukawa, 2003). Acid soils also disturb the existence and persistence of soil rhizobia, affecting the efficacy of nodulation and N fixation in legume species (Graham and Vance, 2003).

Wheat *(Triticum aestivum L.)* belongs to the family Poaceae (Sagar et al., 2018) and it is the world's most important cereal crop in terms of both area cultivated (232 million ha) and amount of grain produced (595 million tons). It is the second most important cereal crop in Bangladesh next to rice; the country still imports significant quantities of wheat to meet the domestic demand (Barma et al., 2019). One of the major factors leading to the decrease in wheat yield in Bangladesh is aluminium toxicity and unavailability of aluminium tolerant cultivars. However, the development of tolerant cultivars is time consuming and laborious. Another strategy to recover the yield limitation is the amelioration of Al toxicity by using different plant hormones and different growth inducing agents. Boron is one of the vital microelement for plant development (Tammam et al., 2012) and is proposed in many plants to reduce the toxicity of Al. Boron supplementation to Al stressed plants increases root elongation in *Citrus grandis* (Zhou et al., 2015), pea (Li et al., 2017), rape seedlings (Yan et al., 2018b), and trifoliate orange (Riaz et al., 2018).

Furthermore, B can alleviate Al toxicity by reducing $Al^{3+}$ binding sites (carboxyl groups) in cell wall. Moreover, increased B application could reduce root oxidative damage by changing cell wall composition and structure (Yan et al., 2018). Zhu et al. (2019) pointed out that B alleviates Al toxicity by altering pectin content and its properties. Moreover, B could enhance Al transfer from the cytoplasm to vacuoles by regulating Al-related transporters in rice root (Zhu et al., 2019). However, the identification of wheat cultivars which are tolerant to aluminium toxicity is of particular significance for their immediate use in regions where aluminium toxicity limit yield. There are few reports on the screening of cultivars tolerant to aluminium toxicity among the wheat cultivars commonly grown in Bangladesh. Besides this, the alleviation of Al toxicity by B in wheat is still in its infancy and least understood. Therefore, in the current experiment, we attempted to determine whether B under Al toxicity conditions may provide an effective strategy for combating the toxicant stress and try attempting to determine how B-Al interaction affects the growth, morphology and physiological performance in wheat, as is the case for many other food crops grown under diverse environmental stress conditions.

## Materials and Methods

The amelioration of Al toxicity of wheat genotypes with B supplementation were evaluated at two stages of wheat for instance, germination stage in petri dish and seedling stage in hydroponics. Both the experiments were conducted at Plant Physiology Laboratory, Department of Crop Botany, Bangladesh Agricultural University, Mymensingh during the period from January, 2016 to February, 2017. Five varieties of wheat were grown (BARI Gom-23, BARI Gom-24, BARI Gom-28, BARI Gom-27, BARI Gom-30) with four treatment combinations (T1 = 0 µM B + 0 µM Al, T2 = 0 µM Al + 40 µM B, T3 = 200 µM Al + 40 µM B and T4 = 0 µM B + 200 µM Al). All of the wheat varieties were collected from the research institute namely Bangladesh Agricultural Research Institute (BARI).

For the germination experiment, seeds of the wheat varieties were sterilized with 5% sodium hypochlorite and then rinsed with distilled water (DW). The two factorial experiment was laid out in a Completely Randomized Design (CRD) with three replications. Hence, sixty petri dishes (5-variety × 4 treatments × 3 replication) with filter papers were set up. Ten seeds of each variety were placed in the petri dish for germination. The Al and B treatments were applied to the petri dish twice a day to ensure the wet environment for seed germination. Germination percentage, vigor index, radicle and plumule length were studied at 4, 6, 8 days after sowing (DAS). Besides these, stress tolerance indices of these variables were also evaluated at 8 DAS using formula stated in Sagar et al., (2019; 2020) and Huqe (2021).

$$Vigor\ Index = Seedling\ length \times \%\ germination$$

$$Stress\ Tolerance\ Index = \frac{Data\ at\ stress}{Data\ at\ control} \times 100$$

The hydroponic experiment was carried out in the growth chamber inside the Plant Physiology Laboratory, Department of Crop Botany, Bangladesh Agricultural University, Mymensingh, Bangladesh, in 2017. The hydroponic system was maintained in plastic pot (8L). Perforated styrofoam sheets were used as trays having 4 holes per tray in each pot. Pregerminated seven-day-old seedlings of all cultivars were placed in the holes of the pot using a piece of Styrofoam to fix the young plants. After seven days, the seedlings were allowed to grow on modified Hoagland´s nutrient solution (pH 5.5–6) with following composition: $Ca(NO_3)_2.4H_2O$ (2 mM), $KH_2PO_4$ (0.2 mM), $K_2SO_4$ (1 mM), $CaCl_2.2H_2O$ (2 mM), $MgSO_4.7H_2O$ (0.5 mM), Fe-EDTA (200 µM), $H_3BO_3$ (1 µM), $CuSO_4.5H_2O$ (0.3 µM), $MnSO_4.6H_2O$ (2 µM), $(NH_4)_6Mo_7O_{24}$ (0.01 µM), $ZnSO_4.7H_2O$ (0.5 µM). Al and B treatments were imposed after seven days of adding nutrients in the pots and kept for 8 days to observe the Al tolerance. The seedlings of all cultivars were evaluated under four growing conditions: T1 = 0 µM B + 0 µM Al, T2 = 0 µM Al + 40 µM B, T3 = 200 µM Al + 40 µM B and T4 = 0 µM B + 200 µM Al.

The experiment was followed by a completely randomized design (CRD) with three replications. Each pot was accommodated by four hills of each. Therefore, total 60 pots were used. A photoperiod of 16 h was maintained, providing artificial lighting using high-pressure sodium lamps (HPS, 400 Watt) and approximately 350 µmol $m^{-2}$ $s^{-1}$ of Photosynthetic Photon Flux Density (PPFD) was provided for the seedling growth. An optimum growing temperature (30±2°C day and 25±1°C night) was maintained during the experiment. The nutrient solution was replaced once a week and aerated continuously using individual air pump for each tank. For the evaluation of the Al tolerance different morphological traits were observed such as root length, shoot length, total fresh weight and total dry weight at 4, 6, 8 DAS. Besides these, stress tolerance index of these parameters were also measured using the above mentioned formula.

Data were statistically analyzed for analyses of variance (ANOVA) using the M-STAT Statistical Computer Package Programme in accordance with the principles of Completely Randomized Design (Gomez and Gomez, 1984). Duncan's Multiple Range Test (DMRT) was used to compare variations among the treatments.

## Results

***Amelioration effect of B for Al toxicity on germination percentage and vigor index at germination stage*:** The effect of Al and B treatments had significant variations on germination percentage and vigor index (Figure 1). The highest germination percentage and vigor index were recorded in BARI Gom-28 with the treatment 0 µM Al + 40 µM B (90.53% and 1325.04) at 8 DAS whereas lowest germination percentage in BARI Gom-30 (65.26%) and vigor index in BARI Gom-27 (260.15) with 0 µM B + 200 µM Al treatment. It was found that 8 days' exposure of Al stress (200 µM Al alone) decreased significantly the germination percentage and vigor index (Fig. 1) compared to other treatments. At 200 µM Al exposure, the germination percentage and vigor index were highest in BARI Gom-28 followed by BARI Gom-23 and lowest in BARI Gom-27. However, the 40 µM B of in combination with the 200 µM Al exhibited significant amelioration against Al stress and the highest percent increase in germination percentage and vigor index were observed in BARI Gom-27 (45.33% and 107.17%) followed by BARI Gom-24 (30.51% and 76.97%) over 200 µM Al alone.

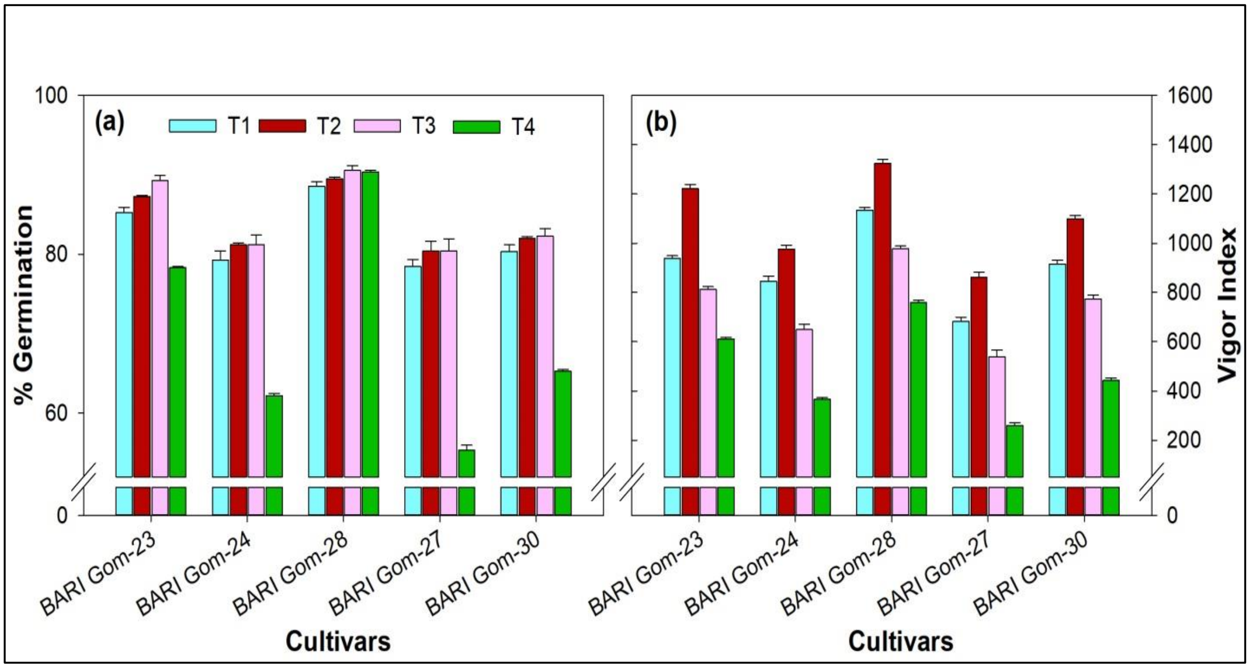

**Figure 1. a)** Germination percentage and **b)** Vigor Index in five wheat cultivars growing in four stress conditions ($T_1$=0 µM B + 0 µM Al, $T_2$=0 µM Al + 40 µM B, $T_3$=200 µM Al + 40 µM B, $T_4$=0 µM B + 200 µM Al) during germination stage. The vertical bars represent the standard error of the means (*n*=3).

***Amelioration effect of B for Al toxicity on radicle and plumule length at germination stage*****:** The effect of Al and B treatments had significant variation on radicle and plumule length of five wheat cultivars (Table 1). The longest radicle length (6.8, 7.7 and 8.6 cm for 4, 6 and 8 DAS respectively) and plumule length (4.8, 5.2 and 6.2 cm for 4, 6 and 8 DAS respectively) were recorded in the treatment combination of BARI Gom-28 with 0 µM Al + 40 µM B. On the other hand, the shortest radicle length (1.8, 2.2 and 2.5 cm for 4, 6 and 8 DAS respectively) and plumule length (1.1, 1.7 and 2.2 cm for 4, 6 and 8 DAS respectively) were observed in BARI Gom-27 with 0 µM B + 200 µM Al. Table 1 depicts that the plants treated with boron in Al stress conditions showed higher radicle and plumule length over alumunium stress alone.

**Table 1.** Effect of aluminium and boron treatment on germination characters of five wheat cultivars at germination stage.

| Treatment | Variety | Radicle length (cm) | | | Plumule length (cm) | | |
|---|---|---|---|---|---|---|---|
| | | 4 DAS | 6 DAS | 8 DAS | 4 DAS | 6 DAS | 8DAS |
| $T_1$ (0 µM B + 0 µM Al) | BARI Gom-23 | 4.7 e | 6.400 e | 6.200 ef | 3.60 f | 3.90 e | 4.80 g |
| | BARI Gom-24 | 3.8 g | 5.400 h | 6.567 de | 2.90 h | 3.60 f | 4.10 j |
| | BARI Gom-28 | 4.8 e | 6.700 d | 7.600 c | 3.80 e | 4.20 d | 5.20 d |
| | BARI Gom-27 | 3.2 i | 4.300 jk | 4.800 h | 2.80 i | 3.30 g | 3.90 k |
| | BARI Gom-30 | 4.3 f | 6.100 f | 6.800 d | 3.30 g | 3.80 e | 4.60 h |
| $T_2$ (0 µM Al + 40 µM B) | BARI Gom-23 | 6.7 a | 7.400 b | 8.200 ab | 4.60 b | 4.90 b | 5.80 b |
| | BARI Gom-24 | 5.8 c | 6.400 e | 6.900 d | 3.90 d | 4.60 c | 5.10 e |
| | BARI Gom-28 | 6.8 a | 7.700 a | 8.600 a | 4.80 a | 5.20 a | 6.20 a |
| | BARI Gom-27 | 5.2 d | 5.300 hi | 5.800 f | 3.80 e | 4.30 d | 4.90 f |
| | BARI Gom-30 | 6.3 b | 7.100 c | 7.800 bc | 4.30 c | 4.80 b | 5.60 c |
| $T_3$ (200 µM Al + 40 µM B) | BARI Gom-23 | 3.7 g | 5.267 hi | 5.300 g | 2.60 j | 2.90 h | 3.80 l |
| | BARI Gom-24 | 2.8 k | 4.400 j | 4.900 gh | 1.90 m | 2.60 i | 3.10 p |
| | BARI Gom-28 | 3.8 g | 5.700 g | 6.600 de | 2.80 i | 3.20 g | 4.20 i |
| | BARI Gom-27 | 2.2 m | 3.300 mn | 3.800 i | 1.80 n | 2.30 j | 2.90 q |
| | BARI Gom-30 | 3.3 hi | 5.100 i | 5.800 f | 2.30 k | 2.80 h | 3.60 m |
| $T_4$ (0 µM B + 200 µM Al) | BARI Gom-23 | 3.0 j | 4.000 l | 4.600 h | 2.10 l | 2.30 j | 3.20 o |
| | BARI Gom-24 | 2.5 l | 3.100 n | 3.600 i | 1.20 p | 1.90 l | 2.30 r |
| | BARI Gom-28 | 3.4 h | 4.100 kl | 4.900 gh | 2.30 k | 2.80 h | 3.50 n |
| | BARI Gom-27 | 1.8 n | 2.200 o | 2.500 j | 1.10 q | 1.70 m | 2.20 s |
| | BARI Gom-30 | 2.8 k | 3.400 m | 3.900 i | 1.70 o | 2.10 k | 2.90 q |
| Level of significance | | * | * | ** | * | ** | * |

In a column, values having similar letter (s) do not differ significantly at 5% level of probability by LSD test. LSD= Least Significant Difference, ** = Significant at 1% level of probability, * = Significant at 5% level of probability.

***Amelioration effect of B for Al toxicity on germination and vigor stress tolerance indices at germination stage*:** The Germination stress tolerance index (GSTI) and Vigor index stress tolerance index (VSTI) exhibited significant variations among combinations of variety and treatment (Figure 2). GSTI and VSTI showed a decreasing pattern with the Al treatment alone compared to the other treatments (Fig. 2). The maximum GSTI and VSTI were observed in BARI Gom-23 with the treatment 40 µM B + 200 µM Al (104.69% and 130.26%) whereas minimum in BARI Gom-27 (70.56% and 38.12%) at 200 µM Al treatment alone. At 200 µM Al exposure, the GSTI and VSTI were the highest in BARI Gom-28 (102.03% and 66.96%) and lowest in BARI Gom-27 (70.56% and 38.12%). In case of, 40 µM B in combination with the 200 µM Al exhibited significant positive response against Al stress and the BARI Gom-27 showed the greatest percent increase (45.33% and 107.17%) in GSTI and VSTI followed by BARI Gom-24 (30.51% and 76.97%) compared to 200 µM Al alone.

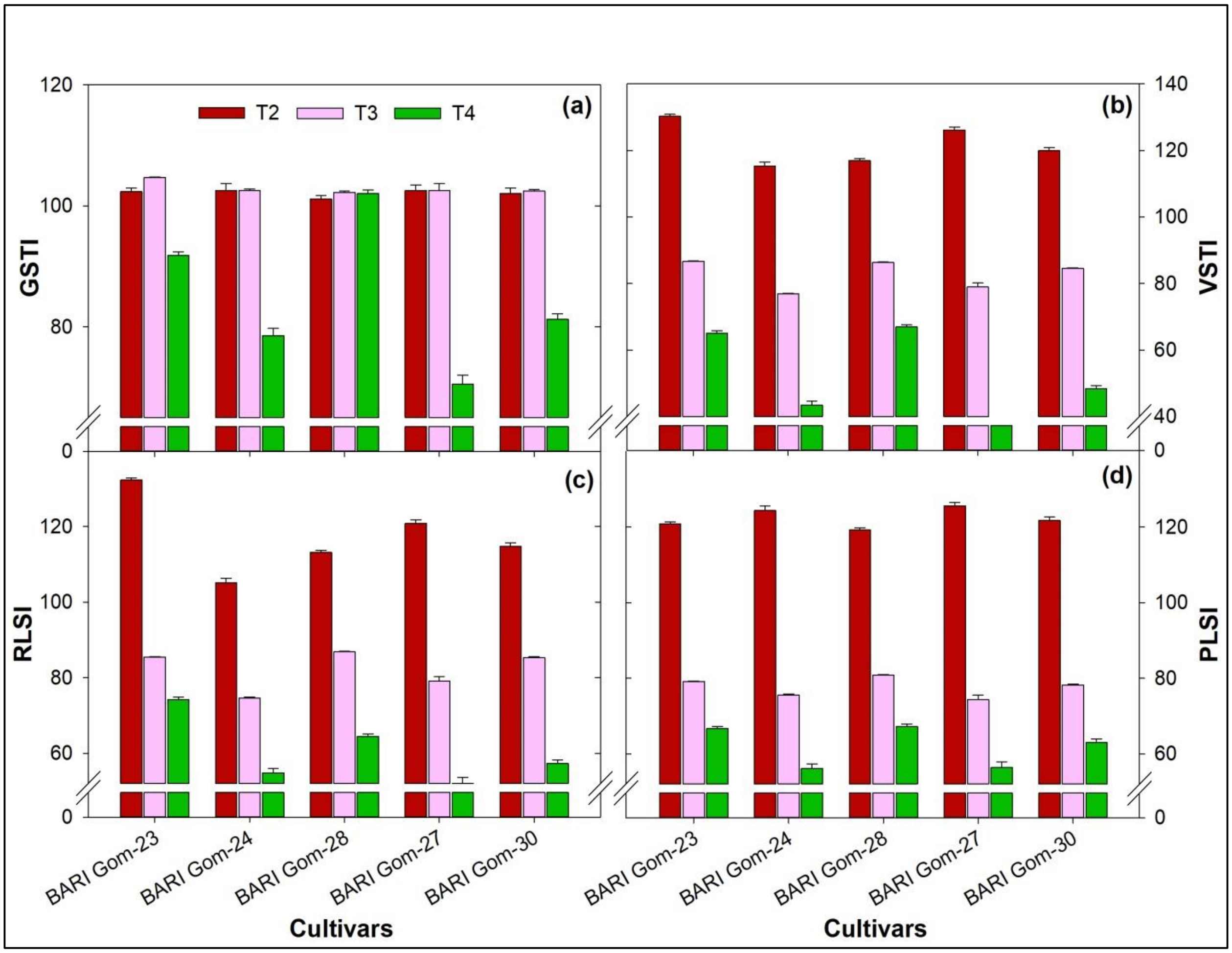

**Figure 2: a)** Germination Stress Tolerance Index (GSTI), **b)** Vigor Stress Tolerance Index (VSTI), **c)** Radicle Length Stress Tolerance Index (RLSI) **d)** Plumule Length Stress Tolerance Index (PLSI) in five wheat cultivars growing in four stress conditions ($T_1$=0 µM B + 0 µM Al, $T_2$=0 µM Al + 40 µM B, $T_3$=200 µM Al + 40 µM B, $T_4$=0 µM B + 200 µM Al) during germination stage at 20 DAS. The vertical bars represent the standard error of the means (*n*=3).

***Amelioration effect of B for Al toxicity on radicle and plumule length stress tolerance indices at germination stage*:** Radicle length stress tolerance index (RLSI) and Plumule length index stress tolerance index (PLSI) reduced significantly with the exposure of Al stress (Figure 2). The treatment 0 µM Al + 40 µM B showed the highest RLSI and PLSI for all the varieties and there was a progressive decrease and minimal results in RLSI and PLSI with the Al treatment for all the varieties (Fig. 2). At 200 µM Al stress level, BARI Gom-28 exhibited the greatest RLSI and PLSI (64.47% and 67.30%) and lowest RLSI in BARI Gom-27 (52.08%), PLSI in BARI Gom-24 (56.09%). A significant increase of RLSI and PLSI was found in BARI Gom-27 (52.0%) and BARI Gom-24 (34.78%) respectively with the treatment 200 µM Al + 40 µM B over the 200 µM Al alone.

### *Ameliration effect of boron for Al toxicity on seedlings characters of wheat grown in hydroponic culture*

**Root length and shoot length:** Root and shoot length reduced significantly with the exposure of Al stress (Figure 3). Results delineate that the root and shoot length were varied significantly among the varieties at different treatments with the passage of time. The root length (9.84, 11.3 and 18.03 cm for 10, 15 and 20 DAS accordingly) and shoot length (7.95, 9.8 and 14.52 cm for 10, 15 and 20 DAS respectively) were the highest in the treatment combination of BARI Gom-28 with 0 µM Al + 40 µM B. On the other hand, the root length (6.07, 8.2 and 8.6 cm for 4, 6 and 8 DAS correspondingly) and shoot length (3.1, 3.9 and 4.7 cm for 10, 15 and 20 DAS respectively) were the lowest in BARI Gom-27 with 0 µM B + 200 µM Al. Fig. 3 also shows that the boron treated plants under Al stress conditions produced longer root and shoot length compared to alumunium stress alone at 10, 15 and 20 DAS.

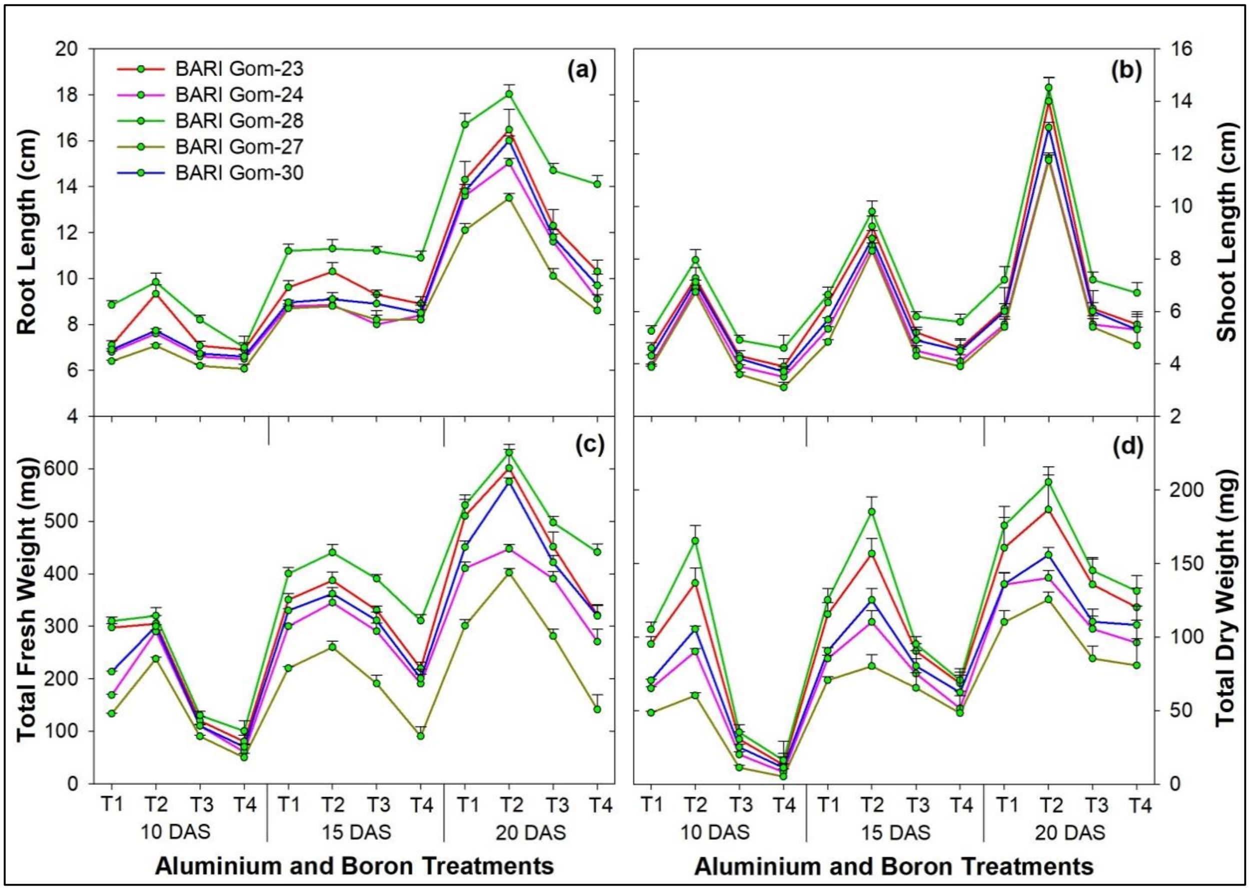

**Figure 3.** Trends of **a)** Root Length (cm) **b)** Shoot Length (cm) **c)** Total Fresh Weight (mg) **d)** Total Dry Weight (mg) in five wheat cultivars growing in four stress conditions ($T_1$=0 µM B + 0 µM Al, $T_2$=0 µM Al + 40 µM B, $T_3$=200 µM Al + 40 µM B, $T_4$=0 µM B + 200 µM Al) in hydroponic culture at 10, 15 and 20 DAS. The vertical bars represent the standard error of the means (*n*=3).

***Amelioration effect of B for Al toxicity on total fresh weight and total dry weight in hydroponic at seedling stage*:** Total fresh weight and total dry weight (mg) were differed significantly among different treatment combinations at 10, 15 and 20 DAS. The total fresh weight (320, 440.3 and 631 mg) and total dry weight (165.5, 185.2 and 205.4 mg) were obtained the maximum value in the treatment combination of BARI

Gom-28 with 0 µM Al + 40 µM B at 10, 15 and 20 DAS respectively. Whereas, total fresh weight (50, 90.67 and 141.5 mg) and total dry weight (5.31, 48.31 and 80.8 mg) were minimum in BARI Gom-27 with Al treated alone at 10, 15 and 20 DAS accordingly. Figure 3 reveals that the boron treated plants under Al stress conditions exhibited larger total fresh and dry weight over alumunium stress alone at 10, 15 and 20 DAS.

***Amelioration effect of B for Al toxicity on root and shoot length stress tolerance indices in hydroponic at seedling stage*:** Root length stress tolerance index (RLSTI) and Shoot length index stress tolerance index (SLSTI) reduced significantly with the exposure of Al stress (Figure 4a,b).

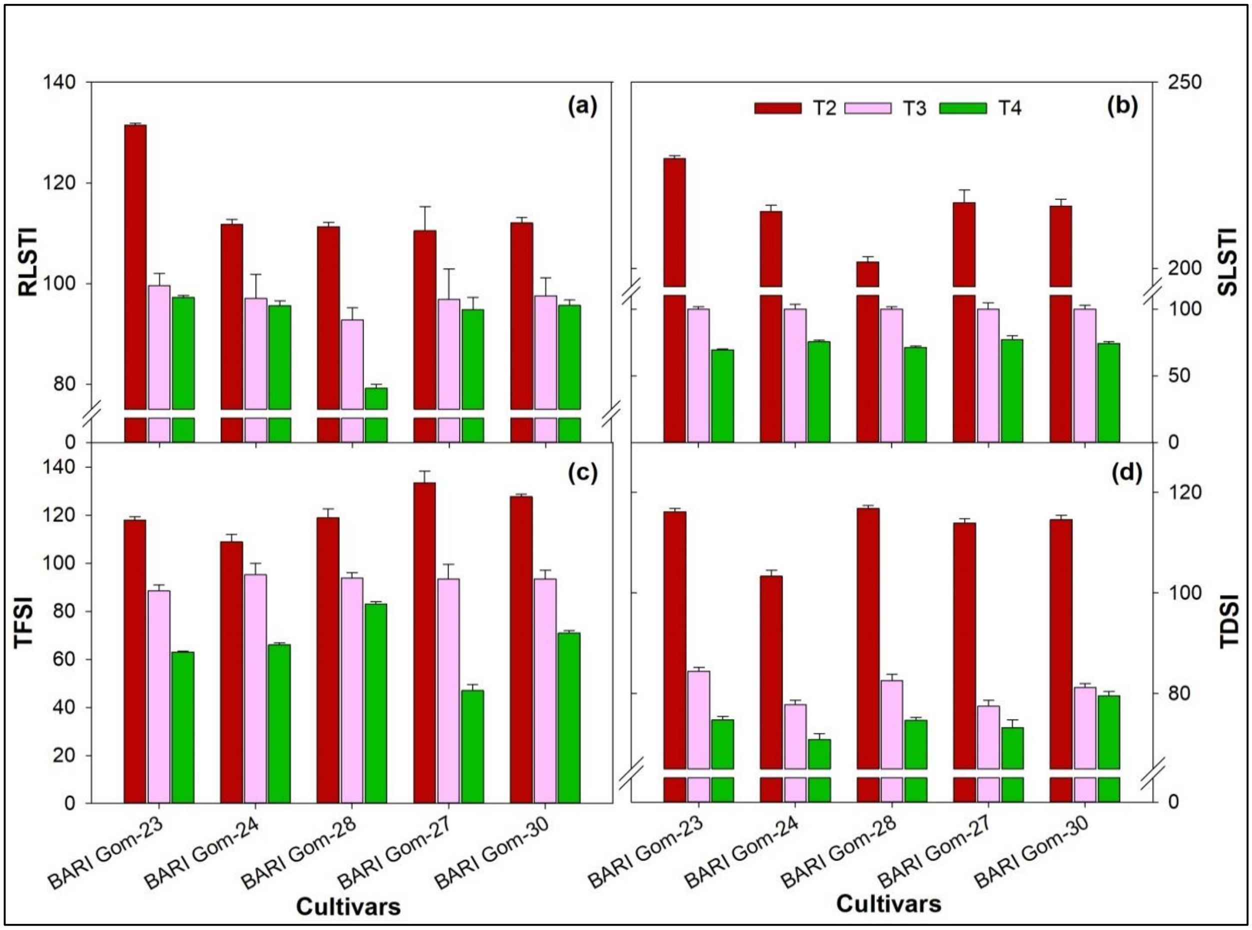

**Figure 4. a)** Root Length Stress Tolerance Index (RLSTI), **b)** Shoot Length Stress Tolerance Index (SLSTI), **c)** Total Fresh Weight Stress Tolerance Index (TFSI) **d)** Total Dry Weight Stress Tolerance Index (TDSI) in five wheat cultivars growing in four stress conditions ($T_1$=0 µM B + 0 µM Al, $T_2$=0 µM Al + 40 µM B, $T_3$=200 µM Al + 40 µM B, $T_4$=0 µM B + 200 µM Al) during seedling stage at 20 DAS. The vertical bars represent the standard error of the means (*n*=3).

The maximum RLSTI and SLSTI was recorded in the treatment 0 µM Al + 40 µM B for all the varieties and there were dramatic reduction and lowest value in RLSI and PLSI with the Al treated conditions (Figure 4a,b). BARI Gom-23 and BARI Gom-27 produced the largest RLSTI (97.18%) and SLSTI (76.92%) respectively whereas lowest RLSTI and SLSTI were found in BARI Gom-28 (79.19%) and BARI Gom-23

(69.49%) accordingly at 200 µM Al alone. Boron (40 µM B) along with 200 µM Al exhibited significantly highest rise of RLSTI and SLSTI was found in BARI Gom-28 (17.14%) and BARI Gom-23 (43.89%) respectively over the 200 µM Al alone.

***Amelioration effect of B for Al toxicity on total fresh and dry weight stress tolerance indices in hydroponic at seedling stage*:** There were significant decreases in Total fresh weight tolerance index (TFSI) and Total dry weight tolerance index (TDSI) with the Al stress (Figure 4c,d). Figure 4c and 4d also revealed that a dramatic reduction and the lowest value in TFSI and TDSI was observed with the Al treated conditions compared to other treatments. At 200 µM Al alone, BARI Gom-28 (83.14%) and BARI Gom-30 (79.57%) displayed the highest TFSI and TDSI whereas BARI Gom-28 (47.01%) and BARI Gom-24 (70.82%) exhibited minimum TFSI and TDSI correspondingly. Boron (40 µM B) responded positively for ameliorating the Al stress and resulted a gradual and maximum increase of TFSI and TDSI in BARI Gom-27 (98.79%) and BARI Gom-23 (12.88%) respectively in comparison with the 200 µM Al alone.

## Discussion

Aluminium ($Al^{3+}$) is found in approximately 40% of the arable soils of the world and acidic soils favor the dissolution of microscopic quantities of $Al^{3+}$ from metal oxides. Aluminium toxicity is a major factor in limiting growth in plants in most strongly acid soils due to several physiological and biochemical pathways. Al toxicity impedes root growth by increasing the reactive oxygen species generation and destroying the homeostasis of antioxidant metabolism (Mora et al., 2006). The initial symptom of Al toxicity is on root growth, leading to changes in root morphology, such as atrophy of root hair, and swelling of root tips (Huang et al., 2014) and decreasing cell wall elasticity and plasticity (Ma et al., 2004; Horst et al., 2010). Thus, considerable effort has been made to cope with the issue in recent years. Since Al stress is a major limitation to plant production on acid soils there is an attempt to develop new cultivars with greater degree of Al tolerance. Therefore, a study was conducted to find out whether the exogenous application of B under Al toxicity conditions may provide an effective strategy for combating the toxicant stress.

The present data showed that 8 days' exposure of Al stress (200 µM Al alone) decreased significantly the germination percentage, vigor index, radicle length and plumule length (Figure 1 and Table 1) compared to other treatments. However, the 40 µM B of in combination with the 200 µM Al exhibited significant amelioration against Al stress and the highest percent increase in germination percentage and vigor index were observed in BARI Gom-27 (45.33% and 107.17%) followed by BARI Gom-24 (30.51% and 76.97%) over 200 µM Al alone. In addition, Table 1 depicts that the plants treated with boron in Al stress conditions showed higher radicle and plumule length over Al stress alone. Moreover, 40 µM B in combination with the 200 µM Al exhibited significant positive response against Al stress and the BARI Gom-27 showed the greatest percent increase (45.33% and 107.17%) in GSTI and VSTI respectively compared to 200 µM Al alone. It has been added that the greater increase in RLSI and PLSI was found in BARI Gom-27 (52.0%) and BARI Gom-24 (34.78%) respectively with the treatment 200 µM Al + 40 µM B over the 200 µM Al alone. The reduction in root elongation with increasing concentrations of Al was recorded in another study (Tamman et al., 2012). It was proposed that the use of supplemental B could protect against root growth inhibition under Al toxicity (Lenoble et al., 1996). Similarly, it was shown that B can ameliorate Al toxicity in mungbean seedlings along with the improvement of root function (Lukaszewski et al., 1996). The results in Experiment 1 to investigate the effect of B on ameliorating the adverse effect of Al treatment on the growth of wheat seedling are in an agreement with the previous studies. Therefore, these Al-toxicity effects were reduced by adequate B supply (40 µM B) in presence of Al. It has been speculated that adequate boron exhibits an antagonistic effect on Al uptake and thus leads to alleviate Al toxicity.

Figure 3 delineates that the root, shoot length, total fresh weight and total dry weight of plant were significantly reduced among the varieties at Al treatment alone with the passage of time. The observed lower values for fresh and dry weight upon Al treatments might be due to aluminium ions were found to affect plasma membrane permeability (Stass and Horst, 1995), fluidity (Gunes et al., 2007) and protein-lipid interactions (Molassiotis et al, 2006). Therefore, these changes under the prevailing experimental conditions caused by Al resulted in a marked disturbance of plasma membrane function and ion transport as well as reduction of water uptake and consequently reduced fresh mass of wheat. Previous studies made by Pereira et al. 2006 shown that the growth, dry weight and fresh weight of roots and shoots of *Cucumber sativus* were decreased at 100, 500, 1000 and 2000 mM $Al_2(SO_4)_3$. Probably, the growth of root cells was affected by aluminium, causing a decrease in cell wall synthesis because aluminium inhibits the secretory function of the Golgi apparatus (Pereira et al., 2006). Figure 3 also shows that the B treated plants under Al stress conditions produced longer root and shoot length; larger total fresh and dry weight compared to alumunium stress alone at 10, 15 and 20 DAS. As a result, different stress tolerance indices like RLSTI, SLSTI, TFSI and TDSI were also higher in plants treated with B under Al stress over Al stress alone.

It was proposed that the use of supplemental B could protect against root growth inhibition under Al toxicity (Lenoble et al., 1996). A study revealed that supplemental B ameliorated Al toxicity in wheat seedlings along with the improvement fresh and dry mass of roots (Tammam et al., 2018). Similarly, it was shown that B could ameliorate Al toxicity in mungbean seedlings along with the improvement of root function (Zhou et al., 2007). In another study showed that high B additions increased epicotyl length of soyabean (Yang et al., 2004) and fresh weight under Al stress, which seems to support the previous reports on B amelioration on Al toxicity (Lenoble et al., 1996). Therefore, the present study also supports the previous studies.

## Conclusion

Wheat yield face a strong challenge due to aluminium toxicity of soil. Similarly, the profound negative effect of Al toxicity on wheat germination and seedling growth has been found in this study. Results revealed that, Al toxicity significantly reduced the germiantiona percentage, vigor index, radicle and plumule length and their stress tolerance indices at germination stage. Furthermore, similar reduction was observed in the root and shoot length and biomass of the wheat cultivars at seedling stage in hydroponics. However, exogenous boron application significantly ameliorated the aluminium toxicity of growth and developments of wheat seedlings in germination and seedling stage. Results indicated that germination percentage, radicle and plumule length, root and shoot length, leaf length, fresh and dry mass plant$^{-1}$ were greater in 0 µM Al + 40 µM B treated plants than 0 µM B + 200 µM Al induced conditions. It indicates that wheat seedlings are susceptible to aluminium and boron can ameliorate aluminium toxicity. Among the studied varieties, BARI Gom-28 had the highest tolerance to aluminium toxicity and positive boron response in respect of growth and development in both germination and seedling stage.

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