

EFFECTS OF ALUMINIUM TOXICITY ON GERMINATION OF SEEDS AND ITS CORRELATION WITH K^+ , Cl^- AND Al^{3+} ACCUMULATION IN RADICLE AND PLUMULE OF *ORYZA SATIVA* L. AND *CICER AERIATINUM* L.

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Keywords: Aluminium toxicity, Germination, K^+ , Cl^- , Al^{3+} accumulation, Plumule, Radicle, Rice, Chickpea

Abstract

Aluminium at concentrations of 10, 50, 100 and 150 μ M inhibited germination of rice and chickpea seeds. Al (10 to 150 μ M) decreased accumulation of K^+ in the radicle and plumule of germinating rice and chickpea seeds from 48 to 96 hrs of treatment. The degree of inhibition increased with the increase in Al concentration. On the other hand, Cl^- accumulation was increased in the radicle and plumule of rice and chickpea seedlings following 10 to 150 μ M Al treatment. A maximum of 2- to 2.4-folds increase in accumulation of Cl^- was observed under Al stress. A 72 hrs exposure to 10 and 100 μ M Al caused a 2.3-folds and 3.8-folds increase in accumulation of Al^{3+} in the radicle and a 1.6- to 2.0-folds increase of that in the plumule of rice seedlings. In the germinating chickpea seeds, Al treatment caused a 2- to 3.4-folds and a 2- to 3-folds increase in accumulation of Al in the radicle and plumule, respectively. Correlation between Al-induced seed germination with K^+ , Cl^- and Al^{3+} accumulation is discussed.

Introduction

Germination potential of seeds is an important factor for growing plants in adverse soil condition like aluminium toxicity. There are a few reports on the effect of aluminium stress on germination of seeds. Al^{3+} decreased seed germination in maize (Nasr 2013). Aluminium at a concentration of 500 ppm had inhibitory effect on wheat seed germination (Alamgir and Akhter 2009). Al toxicity inhibited seed germination in a few plants (Delhaize and Ryan 1995). 50 μ M Al treatment decreased germination percentage in maize (Gumze *et al.* 2007). Al significantly reduced germination of pea (*Pisum sativum* L.) seeds (Singh *et al.* 2011). On the contrary, aluminium toxicity had no effect on germination of wheat (Jamal *et al.* 2006). Significant effect of 50 - 200 μ g Al was not found on germination of tobacco seeds. However, germination time was delayed with increasing Al concentration (Varder *et al.* 2006). There are no reports on the mechanism of aluminium-induced inhibition of seed germination. Accumulation of K^+ , Cl^- and Al^{3+} in plumule and radicle may have some relation with the inhibition of germination of seeds by aluminium.

So in this study, the effect of aluminium on seed germination and its correlation with K^+ , Cl^- and Al^{3+} accumulation in plumule and radicle is reported.

Materials and Methods

Rice (*O. sativa* var. BRRI Dhan-53) and chickpea (*C. arietinum* var. Bari Chhola-7) were taken as experimental plant materials. Seeds of rice were obtained from Bangladesh Rice Research Institute (BRRI) and that of chickpea were procured from Bangladesh Agricultural Research Institute (BARI), respectively.

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Four different concentrations (10, 50, 100 and 150 μM) of AlCl_3 were prepared using half strength Hoagland solution and the pH of each solution was adjusted to 4.2 with 0.2N H_2SO_4 . Half strength Hoagland solution having pH adjusted to 4.2 was used as control.

The seeds were surface sterilized to avoid fungal infection by soaking the seeds with 5.25% sodium hypochlorite for three minutes. The sterilized seeds were submerged in distilled water and aerated for 30 min with an air compressor. Thirty such sterilized seeds were placed on Whatman filter paper contained in a petri dish. Three replicates were used for each treatment. Filter papers were soaked with 10, 50, 100 and 150 μM AlCl_3 (pH 4.2) and half strength Hoagland solution (pH 4.2) was used as control. The chickpea and rice seeds were allowed to germinate in dark at $25^\circ\text{C} \pm 1^\circ\text{C}$ and $30 \pm 1^\circ\text{C}$, respectively. Seeds were considered to be germinated when radicles and plumules could be clearly distinguished. Germination of seeds was recorded at 48, 72 and 96 hrs of Al treatment.

Radicles and plumules of the germinated seeds were separated from cotyledons at 48, 72 and 96 hours from the time of sowing. K^+ and Cl^- were extracted from dry tissue (radicle and plumule) by boiling in a hot water bath following Samad and Karmoker (2013). Al^{3+} was extracted from dry tissue by boiling in a mixture of nitric acid and perchloric acid (4 : 1) using a hot sand bath. Al^{3+} was measured using atomic absorption spectrophotometer (Shimadzu, AA7000, Japan).

Results and Discussion

In rice, aluminium concentration of 10, 50, 100 and 150 μM decreased germination of seeds by 12, 25, 30 and 34%, respectively at 48 hrs of treatment. At 72 hrs of treatment, aluminium (50 - 150 μM) inhibited germination of seeds by 13 to 28%. At 96 hrs of treatment, 50 - 150 μM aluminium decreased germination of rice seeds by 6 to 21% (Table 1).

Table 1. Effects of different concentrations of AlCl_3 on germination of seeds of rice. Each value is the mean of three replicates \pm standard error.

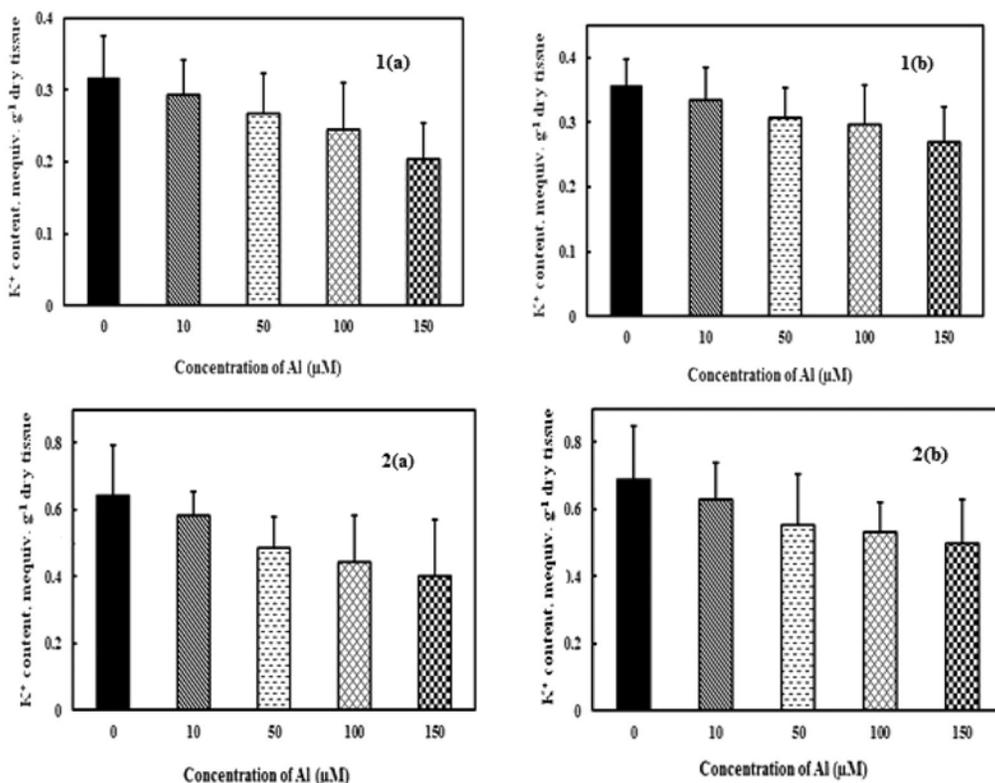
Duration of treatment (hrs)	% germination concentration of AlCl_3 (μM)				
	0	10	50	100	150
48	96 \pm 0.359	88 \pm 0.546	75 \pm 0.333	70 \pm 0.577	66 \pm 0.530
72	100 \pm 0.333	95 \pm 0.571	87 \pm 0.882	79 \pm 0.498	72 \pm 0.672
96	100 \pm 0.333	98 \pm 0.333	94 \pm 0.571	85 \pm 0.667	79 \pm 0.571

In chickpea, aluminium (10 - 150 μM) inhibited seed germination by 20 to 42% at 48 hrs of treatment. At 72 hrs of treatment, 50 - 150 μM aluminium decreased germination of chickpea seeds by 10 to 21%. Aluminium (50 - 150 μM) inhibited seed germination by 7 to 16% at 96 hrs of treatment (Table 2). Aluminium-induced seed germination is supported by the work of Nasr (2013) and Alamgir and Akhter (2009) who recorded inhibition of seed germination of maize and wheat seeds following aluminium treatment.

In rice, accumulation of K^+ in the radicle was decreased by 7% at 10 μM Al treatment and the degree of inhibition increased with the increase in aluminium concentration from 10 - 150 μM and the maximum inhibition was 35% at 150 μM Al at 72 hrs of treatment (Fig. 1a). Similar pattern of inhibition of K^+ accumulation was observed in the plumule of rice following different concentrations of aluminium (10 - 150 μM) treatment at 72 hrs of treatment. The degree of inhibition of K^+ accumulation in the plumule increased with the increase in aluminium concentration from 10 - 150 μM ranging from 6 - 25% (Fig. 1b).

Table 2. Effects of different concentrations of AlCl₃ on germination of seeds of chickpea. Each value is the mean of three replicates \pm standard error.

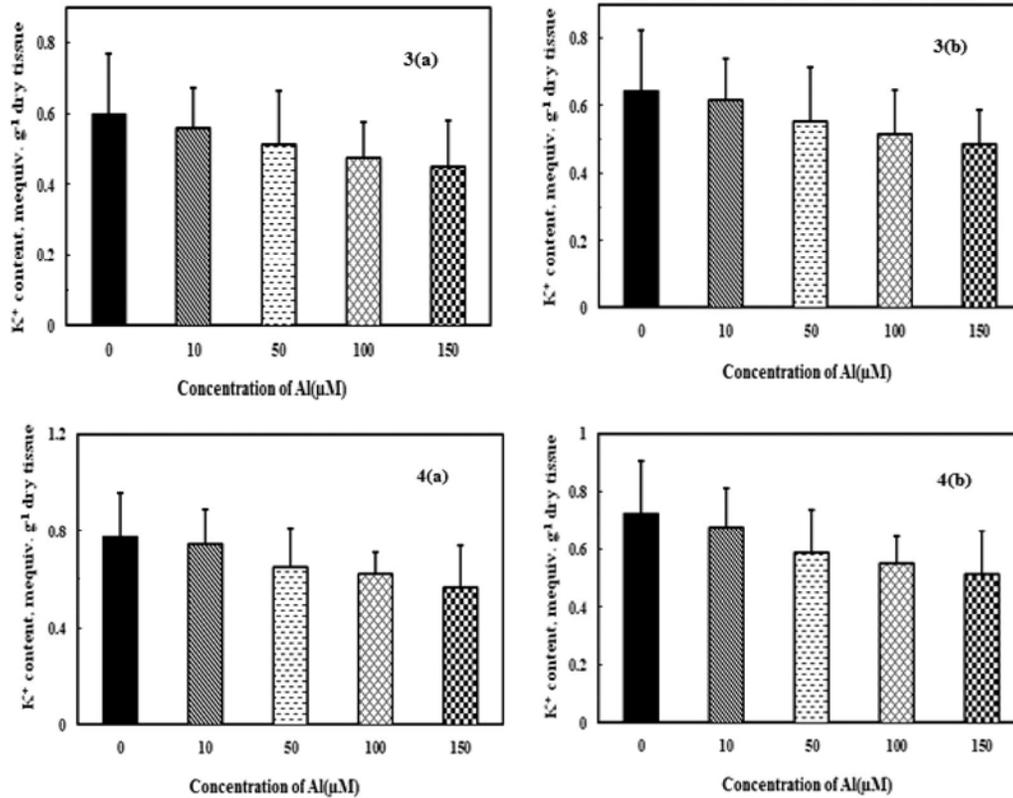
Duration of treatment (hrs)	% germination concentration of AlCl ₃ (μ M)				
	0	10	50	100	150
48	94 \pm 0.577	80 \pm 0.577	72 \pm 0.882	66 \pm 0.946	58 \pm 0.882
72	100 \pm 0.333	94 \pm 0.495	90 \pm 0.651	85 \pm 0.333	79 \pm 0.577
96	100 \pm 0.333	96 \pm 0.568	93 \pm 0.333	89 \pm 0.577	84 \pm 0.333



Figs 1-2: 1. The effect of different concentrations of aluminium (Al) on the accumulation of K⁺ in (a) radicle and (b) plumule of germinating rice seeds at 72 hrs of treatment. ■ represents control; ▨ 10 μ M Al, ▤ 50 μ M Al, ▩ 100 μ M Al and ▧ 150 μ M Al. Each value is the mean of three replicates. Bars represent \pm standard error of the mean value. 2. The effect of different concentrations of aluminium (Al) on the accumulation of K⁺ in (a) radicle and (b) plumule of germinating rice seeds at 96 hrs of treatment. Otherwise as in Fig. 1.

Similarly, 10 - 150 μ M aluminium inhibited K⁺ content in the radicle and in the plumule of rice at 96 hrs of treatment. In this case also the degree of inhibition of K⁺ accumulation in the radicle and plumule increased with the increase in aluminium concentration from 10 - 150 μ M at 96 h of treatment. The inhibition of K⁺ content in the radicle ranged from 9 - 38% and that of K⁺ in the plumule ranged from 9 - 28% at Al concentration ranging from 10 - 150 μ M (Fig. 2a,b).

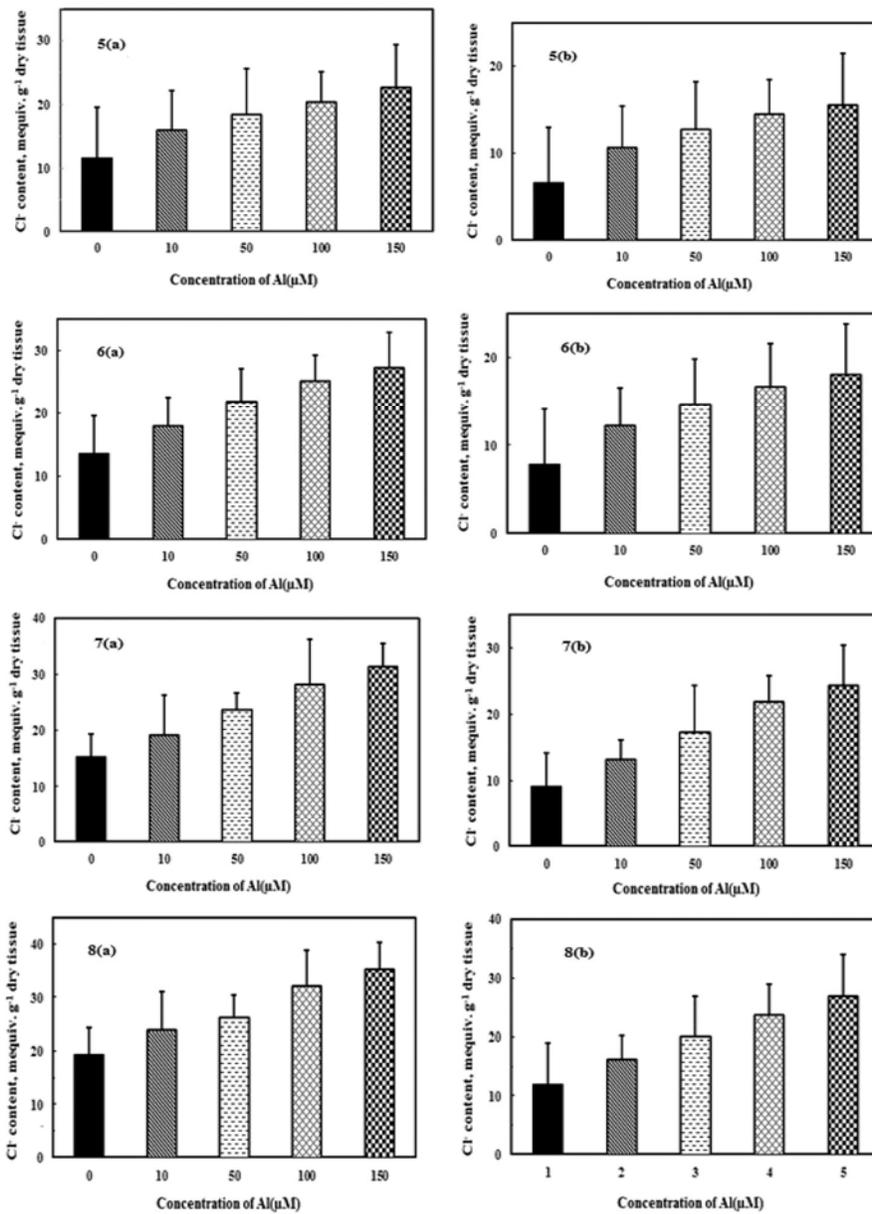
In chickpea, accumulation of K^+ decreased in the radicle by 5 - 24% following 10 - 150 μM aluminium treatment at 72 hrs of treatment (Fig. 3a). Similar magnitude of inhibition of K^+ (4 to 24%) was observed in the plumule of chickpea following Al treatment at 72 hrs of treatment (Fig. 3b).



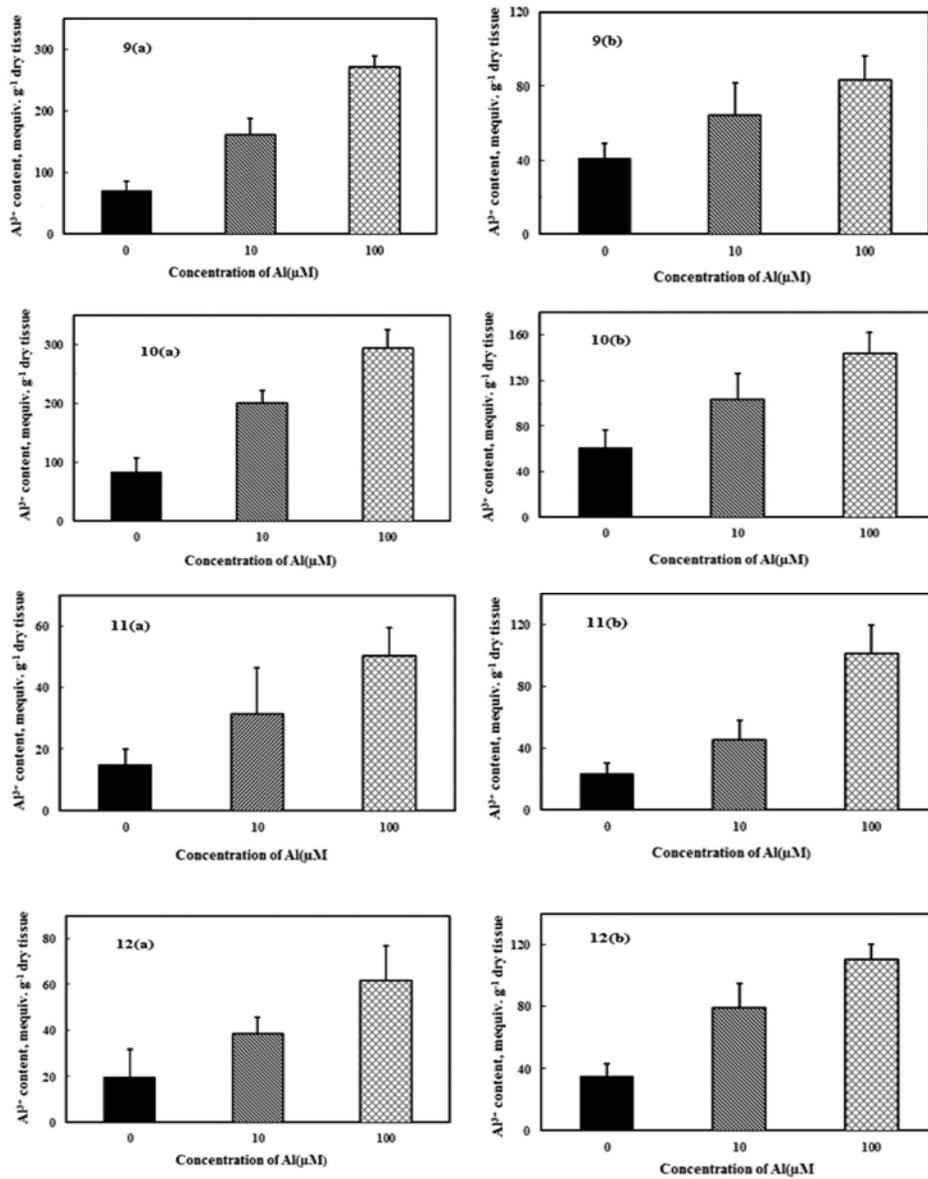
Figs 3-4: 3. The effect of different concentrations of aluminium (Al) on the accumulation of K^+ in (a) radicle and (b) plumule of germinating chickpea seeds at 72 hrs of treatment. Otherwise as Fig. 1. 4. The effect of different concentrations of aluminium (Al) on the accumulation of K^+ in (a) radicle and (b) plumule of germinating chickpea seeds at 96 hrs of treatment. Otherwise as in Fig. 1.

Similarly, 10 - 150 μM aluminium inhibited K^+ content in the radicle and plumule of chickpea at 96 hrs of treatment. The degree of inhibition increased with the increase in aluminium concentration from 10 - 150 μM at 96 hrs of treatment. The inhibition of K^+ content in the radicle of chickpea ranged from 3 - 27% and that of the plumule of chickpea ranged from 6 - 29% at Al concentration ranging from 10 - 150 μM (Fig. 4a, b). This result is supported by Horbowicz *et al.* (2011) who found that high concentration of Al in Hoagland solution decreased K^+ content in cotyledons and hypocotyls of common buckwheat (*Fagopyrum esculentum* Moench).

Aluminium at concentrations of 10, 50, 100 and 150 μM increased accumulation of Cl^- by 38, 60, 77 and 96%, respectively in the radicle of rice at 72 hrs of treatment as compared to control (Fig. 5a). In the plumule of rice, 62% to 2.4-folds increase in Cl^- accumulation was observed at 72 hrs of application of 10 - 150 μM aluminium (Fig. 5b).



Figs 5-8: 5. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating rice seeds at 72 hrs of treatment. Otherwise as in Fig. 1. 6. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating rice seeds at 96 hrs of treatment. Otherwise as in Fig. 1. 7. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating chickpea seeds at 72 hrs of treatment. Otherwise as in Fig. 1. 8. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating chickpea seeds at 96 hrs of treatment. Otherwise as in Fig. 1.



Figs 9-12: 9. The effect of different concentrations of aluminium (Al) on the accumulation of Al³⁺ in (a) radicle and (b) plumule of germinating rice seeds at 72 hrs of treatment ■ represents control, ▨ 10 μM Al ▩ 100 μM Al. Each value is the mean of three replicates. Bars represent ± standard error of the mean value. 10. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating rice seeds at 96 hrs of treatment. Otherwise as in Fig. 9. 11. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating chickpea seeds at 72 hrs of treatment. Otherwise as in Fig. 9. 12. The effect of different concentrations of aluminium (Al) on the accumulation of Cl⁻ in (a) radicle and (b) plumule of germinating chickpea seeds at 96 hrs of treatment. Otherwise as in Fig. 9.

Similarly, 10 - 150 μM aluminium caused a 62% to 2.0-folds increase in Cl^- content in the radicle (Fig. 6a) and a 57% to 2.3-folds increase in the accumulation of Cl^- in the plumule of rice at 96 h of treatment (Fig. 6b).

In chickpea, 10 to 150 μM aluminium caused a 26% to 2-folds increase in Cl^- accumulation in the radicle at 72 hrs of treatment (Fig 7a). In the plumule of chickpea seeds, 10 - 150 μM aluminium caused a 45% - 2.7-folds increase in Cl^- accumulation at 72 hrs of treatment (Fig. 7b).

A 24 to 83% increase in Cl^- accumulation in the radicle was observed at 96 h following 10 and 150 μM aluminium application (Fig. 8a). Similarly, 36% - 2.2-folds increase in Cl^- accumulation in the plumule was observed at 96 hrs following 10 - 150 μM aluminium treatment (Fig. 8b).

At 72 hrs exposure of 10 and 100 μM Al caused 2.3-folds and 3.8-folds increase in accumulation of Al^{3+} , respectively in the radicle of rice (Fig. 9a). Similarly, 10 and 100 μM Al caused a 1.6-folds and 2-folds increase in Al content in the plumule, respectively at 72 hrs of treatment (Fig. 9b).

A 2.4-folds and 3.6-folds increase in Al^{3+} was recorded in the radicle of rice following 10 and 100 μM aluminium, respectively at 96 hrs of treatment (Fig. 10a). In the plumule, 10 and 100 μM Al caused 1.7-folds and 2.4-folds increase in the accumulation of Al^{3+} , respectively at 96 hrs of treatment (Fig. 10b).

Application of 10 and 100 μM Al for 72 hrs caused a 2- folds and 3.4- folds increase in accumulation of Al^{3+} , respectively in the radicle of chickpea (Fig. 11a). In the plumule, exposure of 10 and 100 μM Al for 72 hrs resulted in 2-folds and 4.3-folds increase in Al, respectively (Fig. 11b).

Similarly, a 96 hrs exposure of 10 and 100 μM aluminium increased Al^{3+} accumulation by 2- and 3.2-folds in the radicle of chickpea (Fig. 12a). In the plumule, 10 and 100 μM aluminium treatment caused a 2.2-folds and 3.1-folds increase in accumulation of Al^{3+} , respectively in the plumule at 96 hrs of treatment (Fig. 12b).

Al treatment in the germinating seeds of rice and chickpea decreased K^+ accumulation and increased Cl^- and Al^{3+} accumulation in both the radicle and plumule. It is suggested that aluminium toxicity induced increase in accumulation of Cl^- and Al^{3+} with the concomitant decrease in K^+ accumulation in the radicle and plumule might be responsible for Al-induced inhibition of germination of seeds.

References

- Alamgir ANM and Akhter S 2009. Effects of aluminium (Al^{3+}) on seed germination and seedling growth of wheat (*Triticum aestivum* L.). Bangladesh J. Bot. **38**: 1-6.
- Delhaize E and Ryan PR 1995. Aluminium toxicity and tolerance in plants. Plant Physiol. **107**: 315-327.
- Gumze A, Vinkovic T, Petrovic S, Eded A and Rengel Z 2007. Aluminium toxicity in maize hybrids during germination. Cereal Research Communications. Alps-Adria Scientific Workshop **35**: 421-424.
- Horbowicz M, Kowalczyk W, Grzesiuk A and Mitrus J 2011. Uptake of aluminium and basic elements and accumulation of anthocyanins in seedlings of common buckwheat (*Fagopyrum esculentum* Moench) as a result of increased level of aluminium in nutrient solution. Eco. Chem. Eng. **18**: 479-488.
- Jamal SN, Iqbal MZ and Athar M 2006. Phytotoxic effect of aluminium and chromium on the germination and early growth of wheat (*Triticum aestivum*) varieties Anmol and Kiran. Int. J. Environ. Sci. Tech. **3**: 411-416.
- Nasr N 2013. Germination and seedling growth of maize (*Zea mays* L.) seeds in toxicity of aluminium and nickel. Merit Res. J. Environ. Sci. Toxicol. **1**: 110-113.

- Samad R and Karmoker JL 2013. Effects of NaCl salinity stress on accumulation of K^+ , Na^+ , Cl^- , NO_3^- , sugar and proline contents in the seedlings of Triticale-I. *Bangladesh J. Bot.* **42**: 189-194.
- Singh NB, Yadav K and Amist N 2011. Phytotoxic effect of aluminium on growth and metabolism of *Pisum sativum* L. *Int. J. Innov. Biol. Chem. Sci.* **2**: 10-21.
- Vardar F, Arican E and Gözukirmizi N 2006. Effects of the aluminum on *in vitro* root growth and seed germination of tobacco (*Nicotiana tabacum* L.). *Adv. Food Sci.* **28**: 85-88.

(Manuscript received on 4 March, 2017; revised on 15 May, 2017)