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EFFECTS OF LONG TERM EXPOSURE TO ALUMINIUM STRESS ON THE ACCUMULATION AND DISTRIBUTION OF K^+, Na^+, Ca^{2+}, Mg^{2+}, Fe^{2+} AND Cl^- IN RICE AND CHICKPEA PLANTS

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Key words: Aluminium toxicity, Ion transport, K^+, Na^+, Ca^{2+}, Mg^{2+}, Fe^{2+} and Cl^- accumulation, Rice, Chickpea

Abstract

Sand culture experiments were undertaken to examine the effect of increasing aluminium levels (50-150 μM) on the mineral nutrients uptake (K^+, Na^+, Ca^{2+}, Mg^{2+}, Fe^{2+} and Cl^-). Increasing concentrations of Al inhibited the uptake of K^+, Ca^{2+}, Mg^{2+} and Fe^{2+} but enhanced that of Na^+ and Cl^- in the root and shoot of rice, and the root, stem and leaves of chickpea. 150 μM Al caused a maximum inhibition of K^+ accumulation in the root and shoot of rice ranging from 25.5 to 49.0% and 33 to 55.5%, respectively, from 7 to 28 day of treatment. In the root, stem and leaves of chickpea, 150 μM Al inhibited K^+ content by 23.9 to 84.0%, 13.2 to 54.4% and 25.3 to 61.2%, respectively, from 7 to 28 day of application. On the contrary, a dramatic 2.7 to 3.1-folds and 70.8% to 2.3-folds stimulation of Na^+ accumulation was recorded in the root of rice and chickpea, respectively, following 100 μM Al treatment from 7 to 28 day of treatment. Different concentrations of aluminium led to a stimulation of Cl^- accumulation in different parts of rice and chickpea plants. In rice and chickpea plants, the inhibitory effect of aluminium stress on the accumulation of Ca^{2+}, Mg^{2+} and Fe^{2+} was enhanced with the increase in Al concentration from 50 to 150 μM.

Introduction

Aluminium (Al) is a light metal that makes up 7% of the earth's crust and the third most abundant element after oxygen and silicon. A large amount of Al is incorporated into aluminosilicate soil minerals, and very small quantities appear in the soluble form, capable of influencing biological systems [(1)]. Al is present in all soils, but Al toxicity is manifested only in acid conditions, in which the phytotoxic form Al^{3+} predominates[(2)]. Aluminium (Al) is not regarded as an essential nutrient, but low concentrations can sometimes increase plant growth or induce other desirable effects[(3-5)].

Al ions blocked K^+ uptake in oat[(6)] and in soybean plants[(7)]. Al reduced the absorption of K^+ in four species of coffee[(8)]. On the contrary, Al increased K^+ concentration in *Stylosanthes*[(9)].

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At a concentration of 0.33 mM Al, Na^+ content was highest in maize root but Na^+ concentration decreased above 0.33 mM Al[10].

Al inhibited Ca^{2+} uptake in cultured tobacco cells[11,12]. Ca^{2+} uptake was inhibited by aluminium in root apex of wheat[13,14] and in barley[15]. High aluminium concentration decreased Mg^{2+} accumulation in sorghum[16].

Accumulation of Fe^{2+} was decreased by Al in sorghum[17] and in tomato[18]. Fe^{2+} concentration, in contrast to Ca^{2+}, Mg^{2+} or K^+ was almost four times higher in root and shoot of greek melon under the influence of aluminium[19].

Materials and Methods

Rice (*Oryza sativa* var. BRRI Dhan-53) and chickpea (*C. arietinum* var. Bari chhola-7) were used 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).

Sand culture technique: The seeds were surface sterilized according to Samad and Karmoker[20]. Thirty five surface sterilized seeds were sown in each earthen pot filled with purified quartz sand. The sand was soaked with distilled water and left for germination by covering the pots with black plastic sheet. The pots were kept in net house of Department of Botany, University of Dhaka, under the normal environmental conditions.

The seeds were germinated within 48 hrs of sowing. After germination the plastic sheet was removed from the pots. Rice seedlings were grown in summer at a day/night temperature of 30°C ± 1°C/25°C ± 1°C and day/night length of 14 hrs/10 hrs. Chickpea seedlings were grown in winter at a day/night temperature of 25°C ± 1°C/18°C ± 1°C and day/night length of 10 hrs/14 hrs.

The sand was always moistened with half strength Hoagland solution every 24 hrs. Thinning of the seedlings were done when they were 7-day-old. Seven-day-old seedlings grown in sand culture were subjected to half strength Hoagland solution (pH 4.2) which served as control. Similarly, 50, 100 and 150 μM $AlCl_3$ solution made in half strength Hoagland solution (pH 4.2) were applied to each pot containing 7-day-old seedlings which were used as treatments. Later on, half strength Hoagland solution (pH 4.2) was applied to control plants and 50, 100 and 150 μM $AlCl_3$ solution (pH 4.2) were applied to respective Al-treated plants every day up to 28 days.

Methods of collection of samples from plants: The root and shoot of rice, and the root, stem and leaves of chickpea plants were collected in triplicate at 7, 14, 21 and 28-day of aluminium exposure. Roots were washed in two changes of 0.1 mM $CaSO_4$ to remove free space ions. The samples were dried in an oven at 75°C for 72 hrs. Dry weight of samples was recorded with an electronic balance.

Methods of extraction and analysis of ions from plant samples: K^+, Na^+ and Cl^- were extracted by water digestion using a hot water bath and Ca^{2+}, Mg^{2+} and Fe^{2+} were extracted by digestion in a mixture of nitric and perchloric acid (4 : 1) using a hot sand bath. Potassium (K^+) and sodium (Na^+) ions were measured by flame photometer (Jenway, PEP-7, UK) at a wavelength of 767 nm and 589 nm, respectively. Amount of chloride (Cl^-) ion was measured by standard titrametric method.

The amount of Ca^{2+}, Mg^{2+} and Fe^{2+} in the extract was measured by an atomic absorption spectrophotometer (Perkin-Elmer, Model: A Analyst 200) at wavelengths of 422.67, 285.21 and 248.33 nm, respectively following ASI method.

Rice and chickpea were used as plant materials because reports on the effects of aluminium toxicity on ion transport of these genera are very rare. Here the effect of aluminium toxicity on the accumulation and distribution of K^+, Na^+, Ca^{2+}, Mg^{2+}, Fe^{2+} and Cl^- is reported.

Results and Discussion

Effects of aluminium toxicity on the accumulation and distribution of K^+ in rice and chickpea plants: Amount of 50, 100 and 150 μM Al decreased K^+ accumulation in the root by 7.8 to 27.8%, 15.5 to 36.0% and 25.5 to 49.0%, respectively from 7 to 28 day of treatment (Fig. 1a). A 13.0 to 30.6%, 21.0 to 44.8% and 33 to 55.5% reduction in K^+ content in the shoot of rice plants were observed by 50, 100 and 150 μM Al treatment, respectively from 7 to 28 day of exposure (Fig. 1 b).

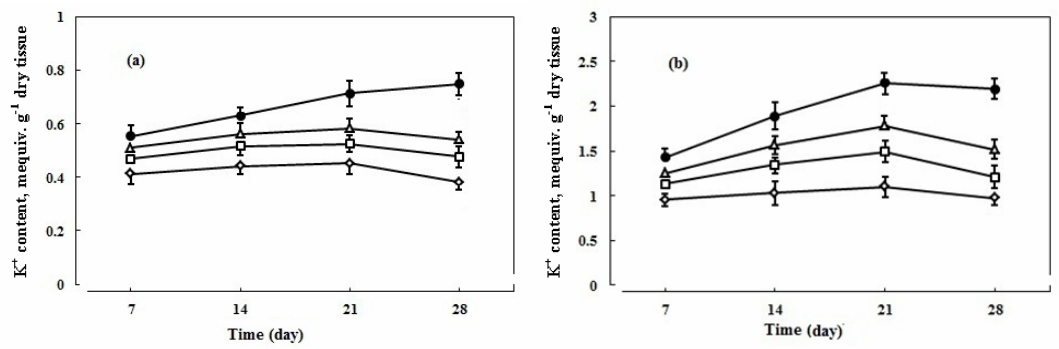

Fig. 1. The effect of different concentrations of aluminium on the accumulation of K^+ in the (a) root and (b) shoot of rice plants grown in sand culture. ● represents control; Δ 50 μM Al; □ 100 μM Al; ◊ 150μM Al. Each value is the mean of three replicates ± standard error.

Amount of 50, 100 and 150 μM Al inhibited K^+ content in the root of chickpea by 15.9 to 61.4%, 19.3 to 75.0% and 23.9 to 84.0%, respectively from 7 to 28 day of application (Fig. 2a). In the stem of chickpea, 50, 100 and 150 μM Al decreased K^+ accumulation by 9.7 to 25.9%, 20.3 to 38.2% and 13.2 to 54.4%, respectively from 7 to 28 day of exposure (Fig. 2b). Accumulation of K^+ in the leaves of chickpea decreased with the increase in Al

concentration from 50 to 150 μM. A maximum of 25.3 to 61.2% inhibition of K^+ in the leaves was caused by 150 μM Al from 7 to 28 day of treatment (Fig. 2c). This result is supported by Bhalerao and Prabhu (2013)[21] who found that Al reduced K^+ uptake in maize and sorghum.

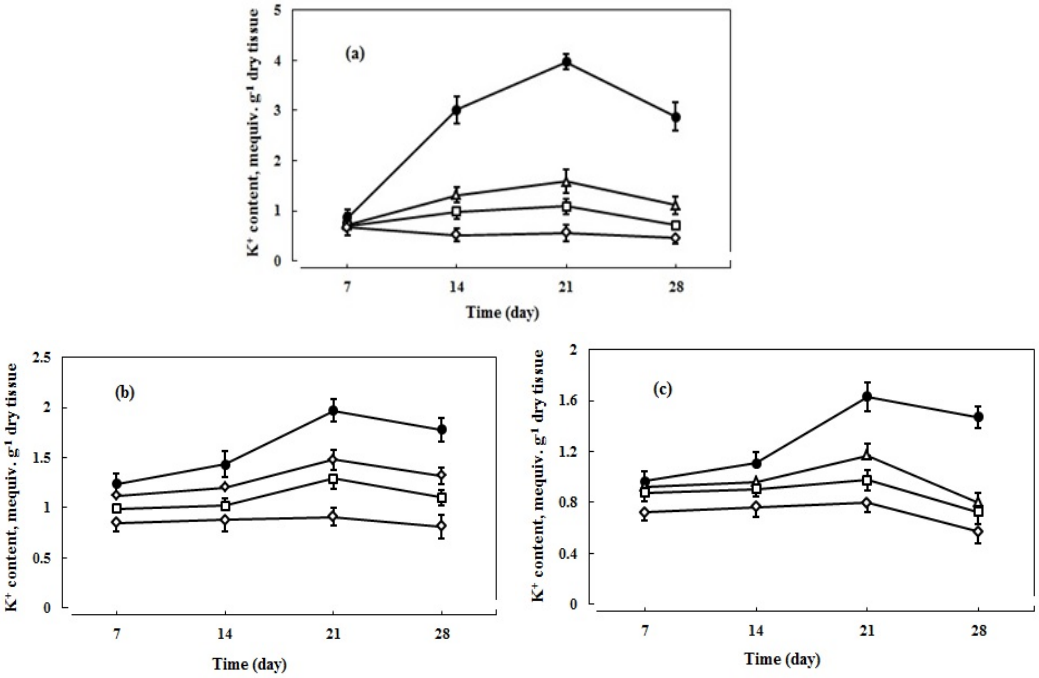

Fig. 2. The effect of different concentrations of aluminium on the accumulation of K^+ in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

Effects of aluminium toxicity on the accumulation and distribution of Na^+ in rice and chickpea plants: In the root of rice, a dramatic 3.2 to 3.7-folds stimulation of Na^+ accumulation was recorded following 150 μM Al treatment from 7 to 28 day of treatment (Fig. 3a). Maximum increase in Na^+ accumulation in the shoot was observed at 150 μM Al ranging from 97.6% to 2.3-folds from 7 to 28 day of treatment (Fig. 3b).

In chickpea plants, 50 and 100 μM Al increased accumulation of Na^+ in the root by 46.9 to 70.3% and 70.8% to 2.3-folds, respectively from 7 to 28 day of treatment. 150 μM Al caused the maximum 2- to 2.4-folds stimulation of Na^+ accumulation in the root from 7 to 28 day of exposure (Fig. 4a). Al, at a concentration of 50, 100 and 150 μM, caused a 31.0 to 64.4%, 66.2 to 94.9% and 85.9% to 2.3-folds increase in Na^+ content, respectively, in the stem of chickpea from 7 to 28 day of application (Fig. 4b). In the leaves of chickpea, 50, 100 and 150 μM Al enhanced Na^+ uptake by 38.3 to 57.3%, 66.7 to 93.8% and 90.0% to 2-folds, respectively from 7 to 28 days of application (Fig. 4c).

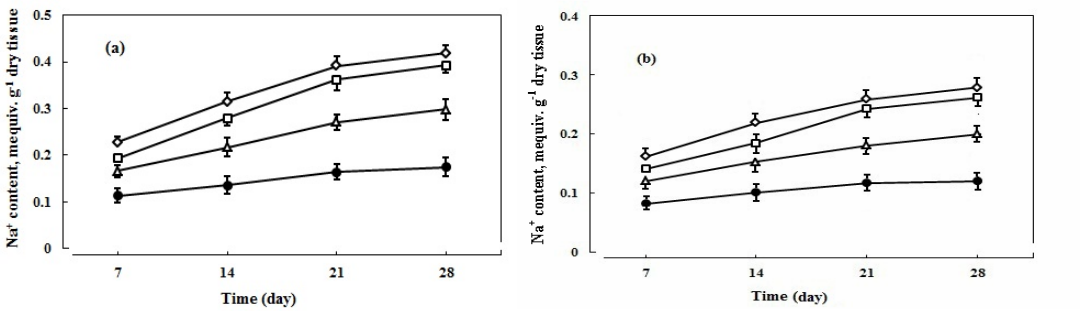

Fig. 3. The effect of different concentrations of aluminium on the accumulation of Na^+ in the (a) root and (b) shoot of rice plants grown in sand culture. Otherwise as Fig. 1.

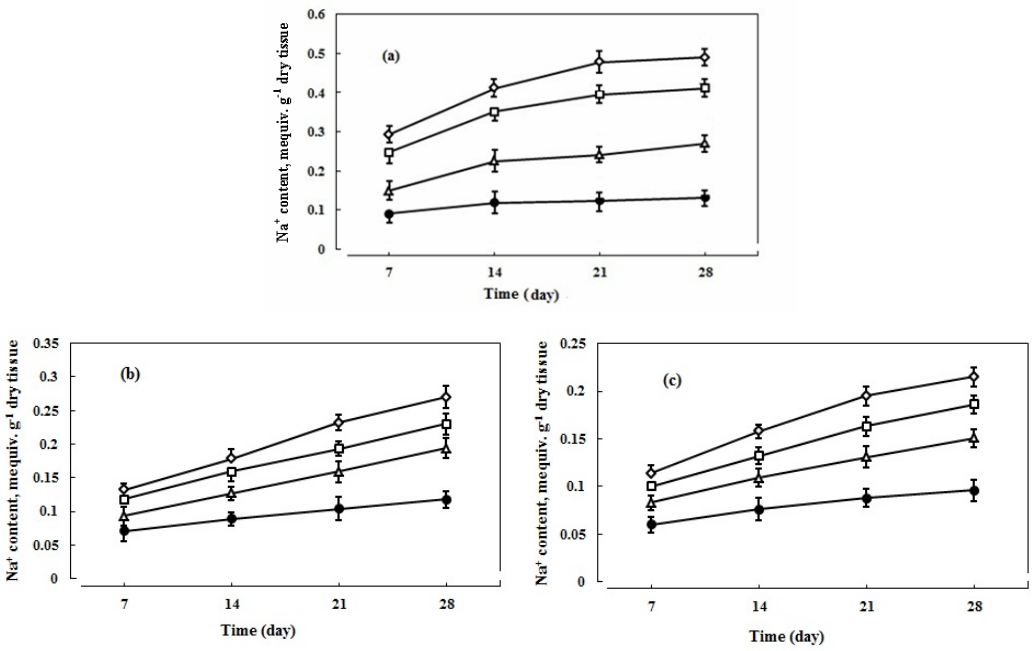

Fig. 4. The effect of different concentrations of aluminium on the accumulation of Na^+ in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

Effects of aluminium toxicity on the accumulation and distribution of Cl^- in rice and chickpea plants: 50, 100 and 150 μM Al stimulated Cl^- accumulation in the root of rice plants from 31.5 to 49.0%, 58.0 to 74.0% and 80.8 to 87.0%, respectively from 7 to 28 day of treatment (Fig. 5a). In the shoot of rice plants, 50 μM Al increased Cl^- accumulation by 38.7% to 2-fold from 7 to 28 day of treatment. 100 μM Al caused a 64.9% to 2.1-fold stimulation of Cl^- content from 7 to 28 day of application. Similarly, 150 μM Al caused the highest 82.0% to 2.3-folds increase in Cl^- accumulation in the shoot from 7 to 28 day of treatment (Fig. 5b).

Different concentrations of Al (50, 100 and 150 μM) increased the accumulation of Cl^- in the root of chickpea plants. 50, 100 and 150 μM Al increased Cl^- accumulation in the root by 21.8 to 96.8%, 1.5 to 2.3-folds and 2 to 2.7-folds, respectively from 7 to 28 day of treatment (Fig. 6a). 50, 100 and 150 μM Al caused 22.5 to 48.3%, 52.8 to 78.0% and 86.0% to 2-folds increase in Cl^- accumulation in the stem, respectively from 7 to 28 day of application (Fig. 6b). Different concentrations of Al (50-150 μM) progressively increased accumulation of Cl^- in the leaves of chickpea. A 20.5 to 82%, 60.2% to 2.3- folds and 97% to 2.7-fold stimulation in Cl^- accumulation in the leaves of chickpea were observed by 50, 100 and 150 μM Al treatment, respectively from 7 to 28 day of application (Fig. 6c). On the contrary, Al decreased accumulation of Cl^- in maize[22].

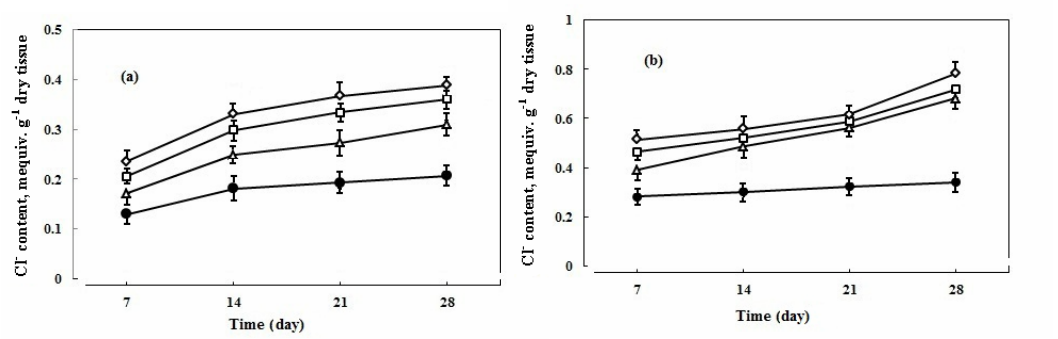

Fig. 5. The effect of different concentrations of aluminium on the accumulation of Cl^- in the (a) root and (b) shoot of rice plants grown in sand culture. Otherwise as Fig. 1.

Effects of aluminium toxicity on Ca^{2+} accumulation in the root of rice and chickpea plants: Fifty μM Al progressively decreased accumulation of Ca^{2+} in the root of rice from 7 to 28 days of treatment. A 35.0 to 70.0% inhibition of Ca^{2+} content was observed in the root of rice exposed to 100 μM Al from 7 to 28 day of exposure. 150 μM Al caused a 61.0 to 74.0% reduction in Ca^{2+} content in the root from 7 to 28 day of application (Fig. 7a). Similarly, in the shoot of rice plants, 50, 100 and 150 μM Al decreased Ca^{2+} content by 15.8 to 41.9%, 29.5 to 52.9% and 34.0 to 60.0%, respectively, from 7 to 28 day of treatment. (Fig. 7b).

In chickpea plants, 50, 100 and 150 μM Al resulted in an inhibition of Ca^{2+} accumulation by 28.5 to 51.5%, 35.8 to 60.2% and 57.0 to 66.8% in the root, respectively, from 7 to 28 day of application (Fig. 8a). In the stem of chickpea, 50, 100 and 150 μM Al caused an inhibition of Ca^{2+} accumulation by 33.0 to 52.8%, 53.9 to 40.0% and 61.5 to 70.4%, respectively from 7 to 28 day of application (Fig. 8b). Al (50 μM) decreased Ca^{2+} accumulation by 29.0 to 51.0% in the leaves from 7 to 28 day of treatment (Fig. 8c). 100 and 150 μM Al caused a 39.4 to 60.3% and 64.6 to 77% inhibition of Ca^{2+}, respectively, in the leaves from 7 to 28 day of application. This result is in agreement with the work of

Zheng *et al.* (2005)[23] who found that Ca^{2+} accumulation decreased progressively in the root of buckwheat with the increase in Al concentrations.

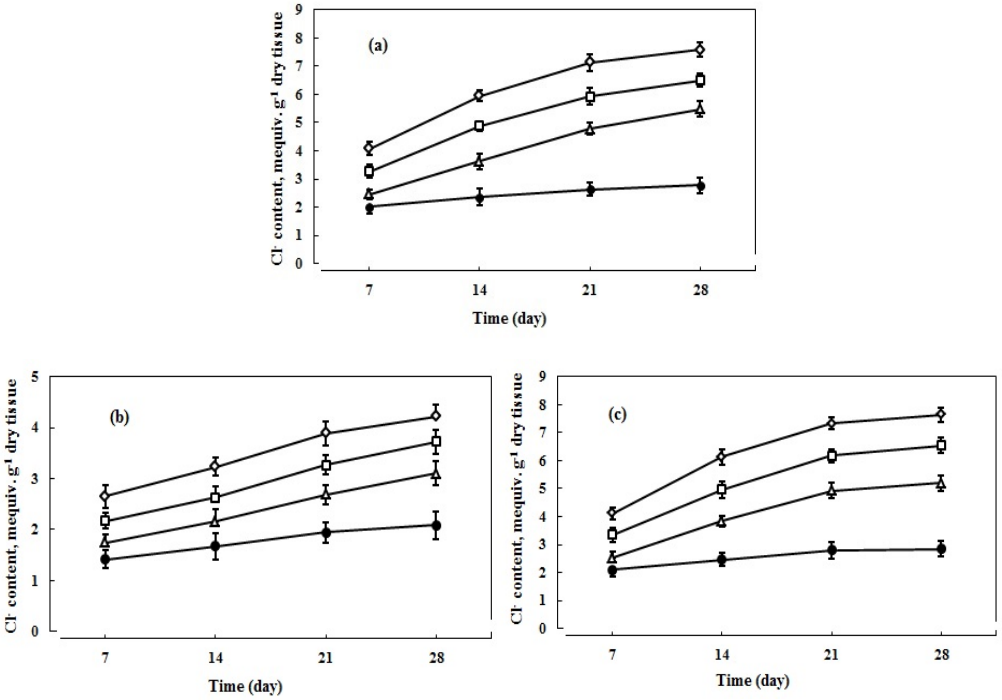

Fig. 6. The effect of different concentrations of aluminium on the accumulation of Cl^- in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

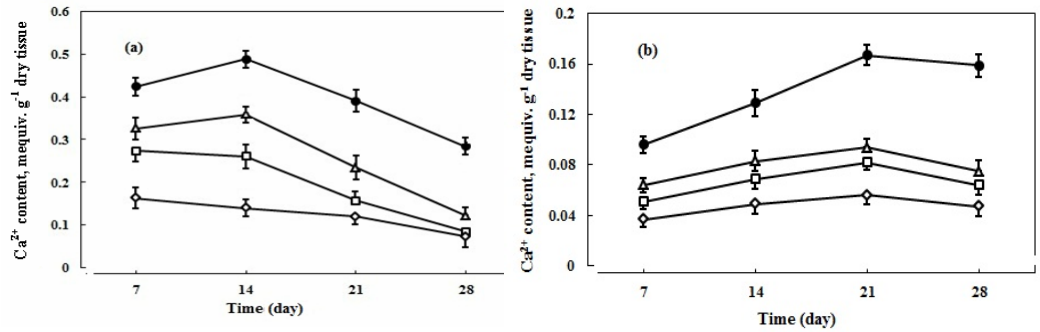

Fig. 7. The effect of different concentrations of aluminium on the accumulation of Ca^{2+} in the (a) root and (b) shoot of rice plants grown in sand culture. Otherwise as Fig. 1.

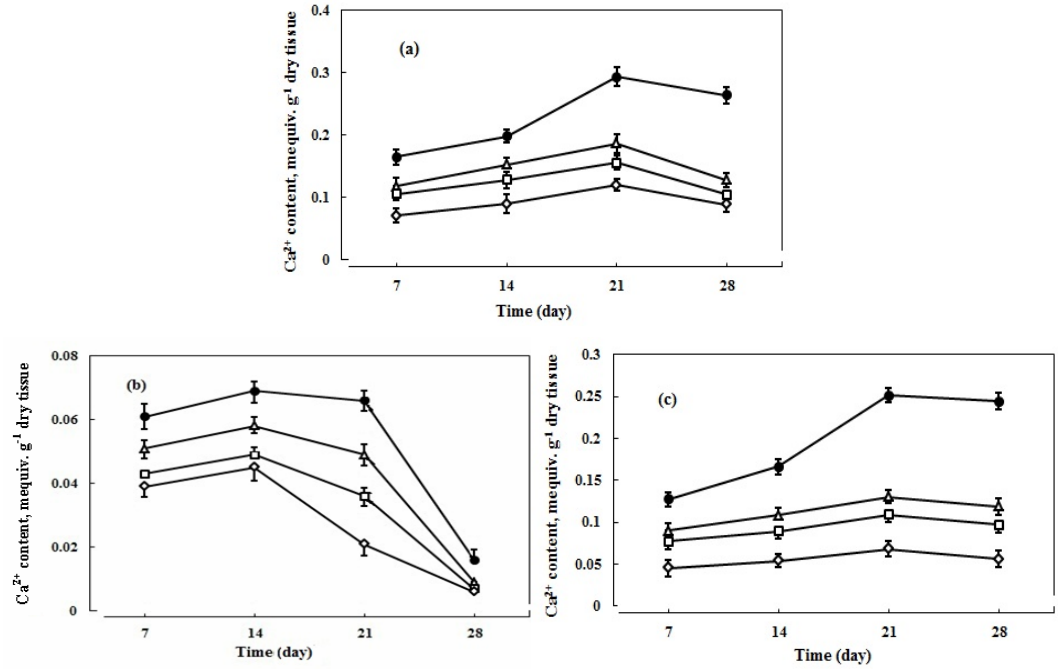

Fig. 8. The effect of different concentrations of aluminium on the accumulation of Ca^{2+} in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

Effects of aluminium toxicity on accumulation and distribution of Mg^{2+} in rice and chickpea plants: Aluminium, at concentrations of 50 and 100 μM, decreased Mg^{2+} accumulation in the root of rice by 28.0 to 56.7% and 39.8 to 66.0%, respectively, from 7 to 28 day of treatment. 150 μM Al caused the highest 52.0 to 75.0% inhibition of Mg^{2+} accumulation in the root from 7 to 28 day of treatment (Fig. 9a). In the shoot of rice, the inhibitory effect increased with the increase in Al concentration from 50 to 150 μM. (Fig. 9b).

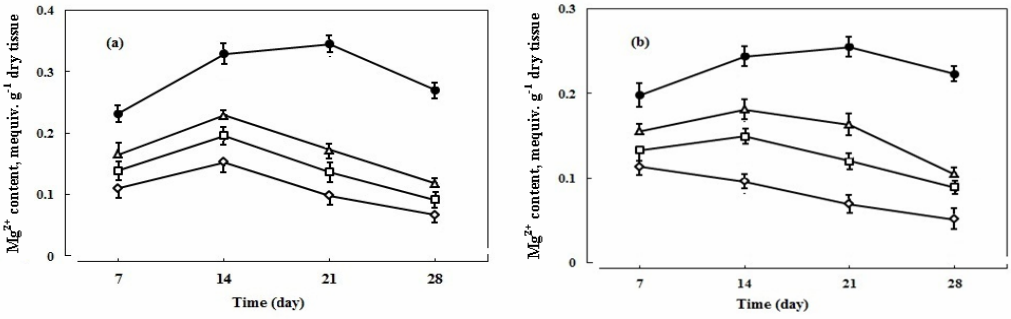

Fig. 9. The effect of different concentrations of aluminium on the accumulation of Mg^{2+} in the (a) root and (b) shoot of rice plants grown in sand culture. Otherwise as Fig. 1.

In chickpea plants, 50, 100 and 150 μM Al decreased the accumulation of Mg^{2+} by 19.3 to 47.9%, 63.7 to 72.8% and 44.4 to 88.8% in the root, respectively from 7 to 28 day of treatment (Fig. 10a). In the stem, the degree of inhibition increased with the increase in concentration of Al from 50 to 150 μM (Fig. 10b). Al, at a concentration of 50, 100 and 150 μM, inhibited the accumulation of Mg^{2+} by 22.0 to 42.8%, 37.7 to 60.6% and 47.7 to 67.6% in the leaves, respectively from 7 to 28 day of treatment (Fig. 10c). Similarly, Al decreased Mg^{2+} accumulation in red spruce[24].

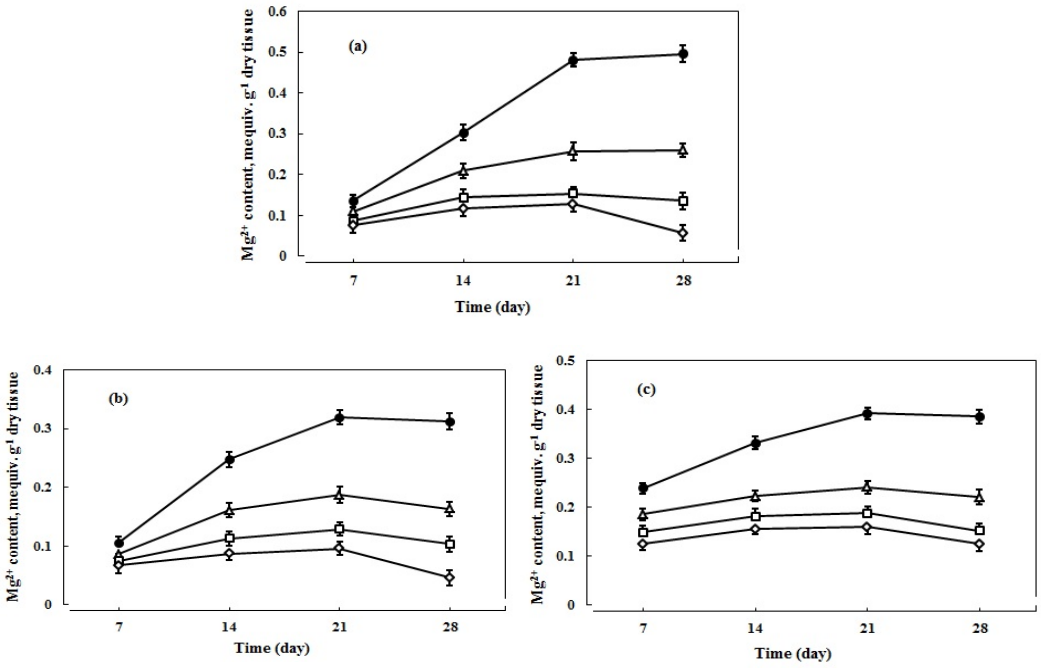

Fig. 10. The effect of different concentrations of aluminium on the accumulation of Mg^{2+} in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

Effects of aluminium toxicity on the accumulation and distribution of Fe^{2+} in rice and chickpea plants: Accumulation of Fe^{2+} in the root of rice progressively decreased from 22.0 to 41.7% and 35.0 to 71.6%, respectively when subjected to 50 and 100 μM Al from 7 to 28 day of treatment. Maximum inhibition of Fe^{2+} accumulation was recorded in the root of rice plants grown in 150 μM Al which ranged from 55.5 to 85.9% from 7 to 28 day of application (Fig. 11a). In shoot of rice plants, a 30.0 to 61.8%, 50.0 to 82.7% and 63.8 to 91.5% inhibition of Fe^{2+} in the shoot was recorded following 50, 100 and 150 μM Al treatment, respectively, from 7 to 28 day of application (Fig. 11b).

In chickpea plants, 50, 100 and 150 μM Al decreased the accumulation of Fe^{2+} in the root by 28.6 to 45.6%, 42.9 to 67.0% and 54.8 to 74.7%, respectively from 7 to 28 day of application (Fig. 12a). In the stem, 50, 100 and 150 μM Al caused a 35.0 to 50.7%, 46.0 to

67.6% and 59.5 to 76.0% inhibition in the accumulation of Fe^{2+}, respectively from 7 to 28 day of application (Fig. 12b). 50, 100 and 150 μM Al inhibited the accumulation of Fe^{2+} in the leaves by 16.0 to 43.6%, 40.0 to 63.6% and 56.0 to 78.0%, respectively, from 7 to 28 day of exposure (Fig. 12c). Similarly, Al stress decreased the accumulation of Fe in sorghum[25].

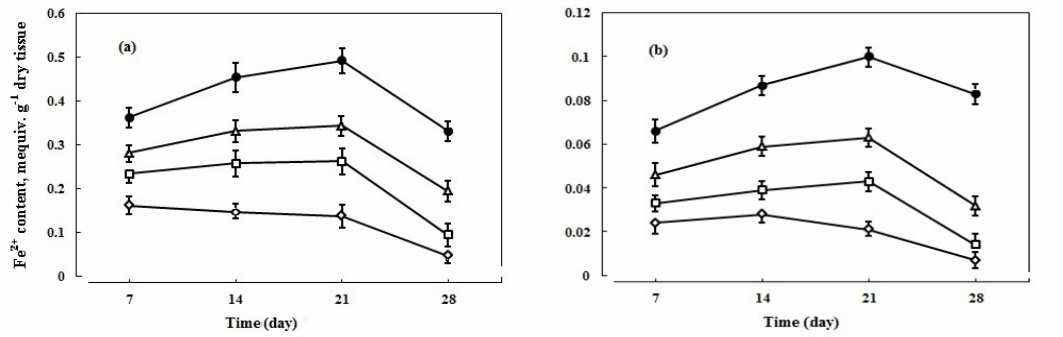

Fig. 11. The effect of different concentrations of aluminium on the accumulation of Fe^{2+} in the (a) root and (b) shoot of rice plants grown in sand culture. Otherwise as Fig. 1.

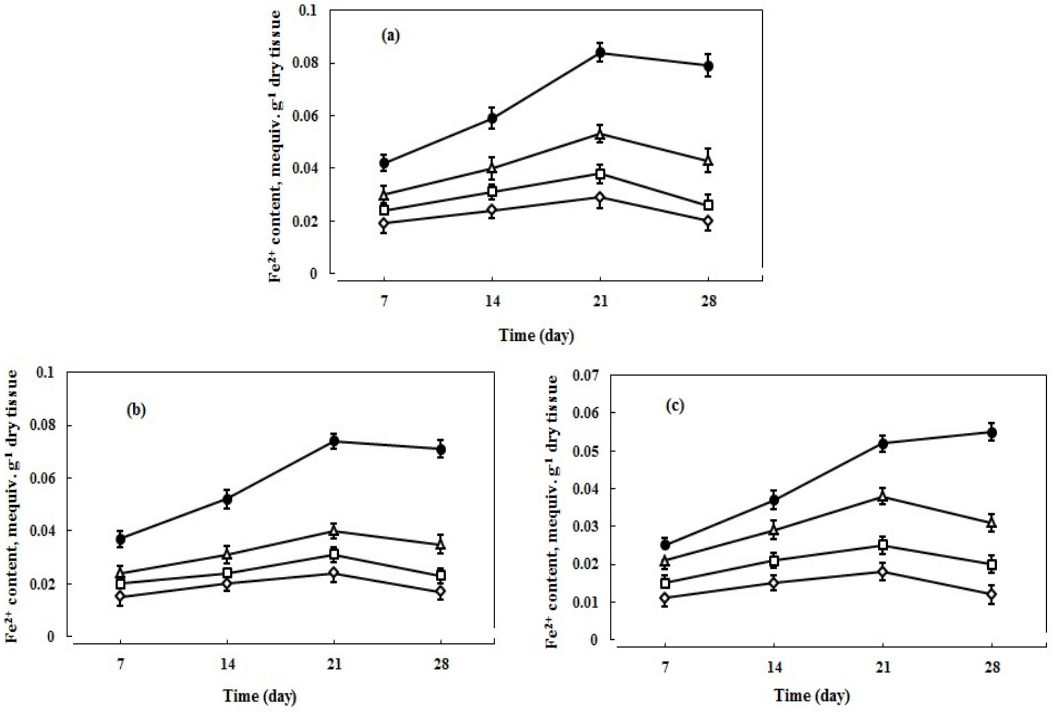

Fig. 12. The effect of different concentrations of aluminium on the accumulation of Fe^{2+} in the (a) root, (b) stem and (c) leaves of chickpea plants grown in sand culture. Otherwise as Fig. 1.

Conclusion: Aluminium toxicity decreased the accumulation of K^+ and increased that in Na^+ and Cl^- in both rice and chickpea plants grown in sand culture. Al-toxicity-induced decrease in K^+ uptake with concomitant increase in that of Na^+ indicates that Al stress alters K^+/Na^+ selectivity.

Al also reduced the accumulation of divalent cations (especially Ca and Mg) by interfering with the membrane transport in rice and chickpea plants grown in sand culture. Iron is the constituent of respiratory enzyme cytochrome oxidase. Therefore, Al-induced decrease in Fe^{2+} accumulation would reduce respiration resulting in a decrease in ion transport which is dependent on respiratory energy.

Thus, aluminium toxicity hinders the uptake of most nutrients thus leading to disturbance of plant growth and development in acid soil.

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