

EFFECTS OF PHOSPHORUS DEFICIENCY ON ION TRANSPORT AND ITS ANATOMICAL RELATION IN MUNG BEAN (*VIGNA RADIATA* L.)

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Abstract

Effects of phosphorus deficiency on ion transport and its relation with anatomical structure in mungbean plants were investigated. Phosphorous deficiency caused an increase in the accumulation of K^+ in the root and a decrease in the stem and leaf of mungbean seedlings grown in solution culture. Na^+ and NO_3^- accumulation was increased in the root, stem and leaf of P-deficient plant. On the contrary, accumulation of PO_4^{3-} was decreased in root, stem and leaf under P-deficiency treatment. The diameter of root was decreased under phosphorus deficiency. Vascular area was found to decrease and occupied less area with smaller size of metaxylem cavity in the root and stem of mungbean plant. In phosphorus deficient plant, phloem elements were poorly developed and occupied smaller area.

Introduction

Mungbean (*Vigna radiata* L.) is an important legume of Asian origin, and is widely cultivated in the countries of Asia, Australia and Africa continents (Yang *et al.* 2008). It is an important summer pulse crop of many South Asian countries including India, Pakistan, Bangladesh, Thailand and Korea (Hussain *et al.* 2006). Phosphorous plays a vital role as a structural component of cell and metabolically active compounds i.e. chloroplasts, mitochondria, phytin, nucleic acid, protein, flavin nucleotides and several enzymes. Phosphorus (P), an essential macronutrient for plant growth, provides indispensable foundation to agricultural production (Nagarjan *et al.* 2011). It is particularly important for the growth of young tissues, flowering and seed formation. It is an important plant macronutrient making up about 0.2% of the plant dry weight. Total soil phosphorus is often hundred fold more than the fraction of inorganic phosphorus available for uptake by crop plants (Sarker and Karmoker 2011). With increasing demand of agricultural production and as the peak in global production will occur in the next decades, phosphorus is receiving more attention as a renewable source (Gilbert 2009). Thus, effects of phosphorus deficiency on the accumulation of Na^+ , K^+ , NO_3^- and PO_4^{3-} and its relation with anatomical structure were investigated.

Materials and Methods

Mungbean (*Vigna radiata* L. var. BARI MUNG-6) was used as plant material. The seeds were collected through the courtesy of Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur. To study the accumulation of different ions, plants were grown in solution culture. Seeds were surface sterilized with 5.25% sodium hypochlorite solution according to Karmoker and Van Steveninck (1979). Plastic lid covered with cotton gauze was placed upon the beaker, and the beakers were painted black to avoid the exposure of light to the roots and filled with nutrient solution, prepared in the laboratory. The nutrient solution contained the following nutrients: K_2SO_4 , KCl, $MgSO_4$, $Ca(NO_3)_2$, KH_2PO_4 and $FeSO_4$ -EDTA. After 48 hrs of sowing, the

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seeds were germinated and the germinated seeds were transferred to light bank at a day/night temperature of $25 \pm 1^\circ\text{C}/18 \pm 1^\circ\text{C}$ and day/night length of 11 hrs/13 hrs and light intensity was $160 \mu\text{E}/\text{m}^2/\text{s}$. The solution was replenished every 48 hrs. The solution was continuously aerated through bubbler with the help of air compressor. Seven day old seedlings were supplied with nutrient solution with or without KH_2PO_4 . Phosphorus-containing solution (+P-solution) was used as control and phosphorus-free solution (-P-solution) was used as phosphorus-deficiency treatment. Samples were collected after 24, 48, 72 and 96 hrs of phosphorus deficiency treatment. Three replicates were used in each treatment.

K^+ , Na^+ and NO_3^- in the root, stem and leaf were extracted by water digestion and PO_4^{3-} was extracted by acid digestion. K^+ and Na^+ were measured using a flame analyzer (Jenway PEP-7, UK) at wavelengths of 767 and 589 nm, respectively. NO_3^- and PO_4^{3-} were measured according to the method of Cataldo *et al.* (1975) and Jackson (1967), respectively.

To study anatomical structures plants were grown in sand culture (Hewitt 1966). Sterilized seeds were sown in pots filled with purified sand. The seeds germinated within 48 hrs of sowing. After germination of seeds, seedlings were supplied with nutrient solution with or without KH_2PO_4 , same as used in solution culture, i.e. sand of one pot was soaked with P-containing solution, control (+P) solution and other pot was soaked with solution lacking of phosphorus, treatment (-P). The sand was moistened with +P or -P solution every 24 hrs. The root and stem of mungbean plants were collected at 45-day of germination. Free hand sectioning was done and the sections were stained with safranin. Transverse sections of the root and stem were studied with the help of a microscope (Nikon ECLIPSE E2000MV R, Japan) and photographs of the sections were taken using a digital microphotography attached to the microscope at different magnification (5 \times , 10 \times and 40 \times).

Results and Discussion

Phosphorus deficiency increased the accumulation of Na^+ in the root by 20 to 22% from 24 to 96 hrs of treatment (Fig. 1a). In the stem, Na^+ accumulation was increased up to 48 hrs of phosphorus deficiency treatment (Fig. 1b). In the leaf, Na^+ accumulation was increased by 10% to 2.8- fold at 24 to 48 hrs and then decreased up to 20% at 96 hrs of treatment (Fig. 1c). Similar increase in Na^+ accumulation occurred for compensation of the decrease in K^+ due to phosphorus deficiency (Nasreen 1999). Phosphorus deficiency increased Na^+ accumulation in the shoot of lentil (Sarker and Karmoker 2011).

Phosphorus deficiency increased K^+ accumulation in the root by 40% to 1.5- fold from 24 to 96 hrs (Fig. 2a) and decreased by 29 to 47% from 24 to 96 hrs of treatment in shoot (Fig. 2b). In the leaf, K^+ uptake showed a reduction under P-deficiency condition (Fig. 2c). Similar increased in K^+ content was observed in the shoot of rape and radish following P-deficiency treatment (Cui *et al.* 2003).

In the root, accumulation of NO_3^- was increased by 80% at 24 hrs of treatment and a stimulatory effect was maintained up to 96 hrs of treatment (Fig. 3a). In the stem, NO_3^- uptake showed an increase under P-deficient condition (Fig. 3b). In the leaf, NO_3^- accumulation increased by 13 to 37% from 24 to 96 hrs of treatment (Fig. 3c). Phosphorus deficiency increased NO_3^- accumulation in the root but decreased that in the shoot of wheat (Mehraz *et al.* 2017). Dinkelaker and Marschner (1992) indicated that, phosphorus deficiency plays an important role in stimulation of N-uptake.

In the root of mungbean seedlings, phosphorus deficiency decreased accumulation of phosphate by 55.77% at 24 hrs of treatment and an inhibitory effect was sustained up to 96 hrs of treatment. In the shoot, PO_4^{3-} accumulation decreased from 24 to 96 hrs of treatment. In the leaf,

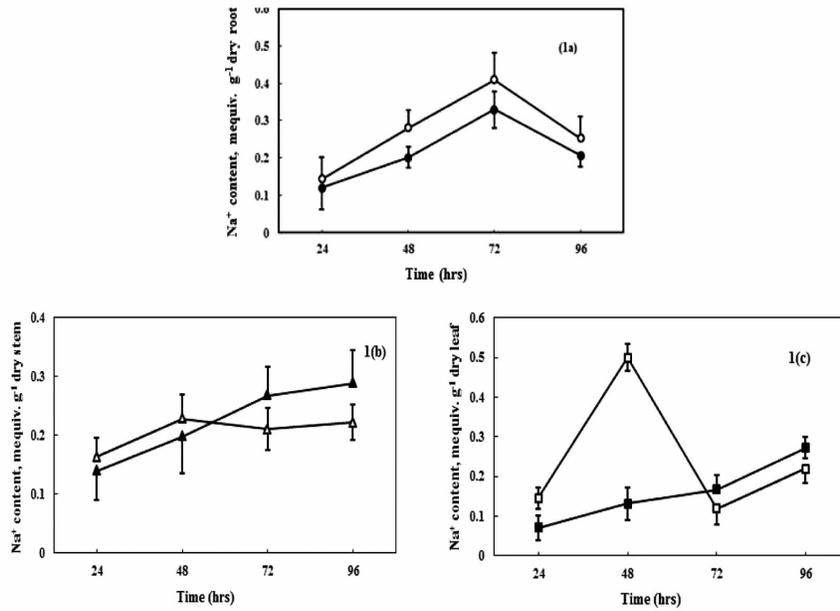


Fig. 1. Effects of Phosphorus deficiency on the accumulation of Na⁺ in (a) root, (b) stem and (c) leaf of mungbean seedlings. Solid symbols (●) represent +P and open symbols (○) represent -P. Each value is the mean of three replicates. Bars represent \pm standard error of the mean value.

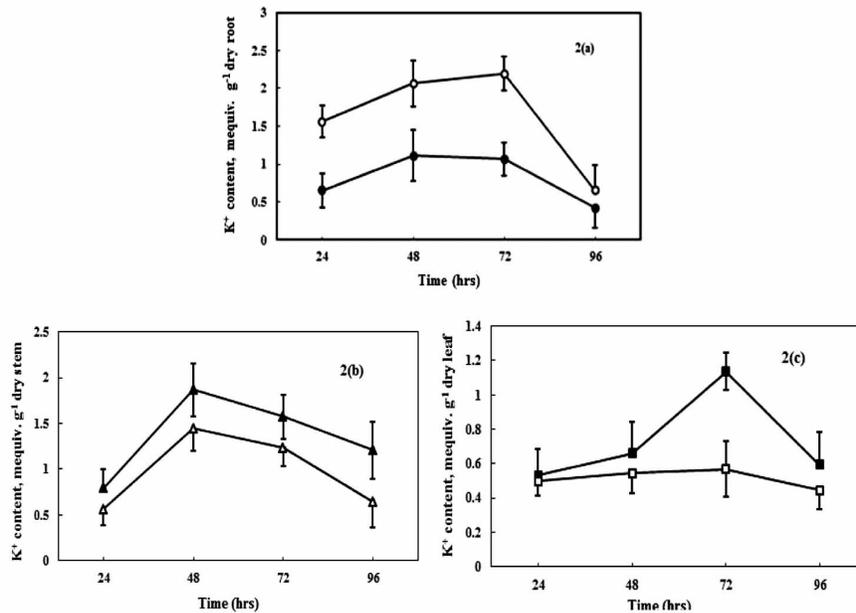


Fig. 2. Effects of Phosphorus deficiency on the accumulation of K⁺ in (a) root, (b) stem and (c) leaf of mungbean seedlings. Otherwise as Fig. 1.

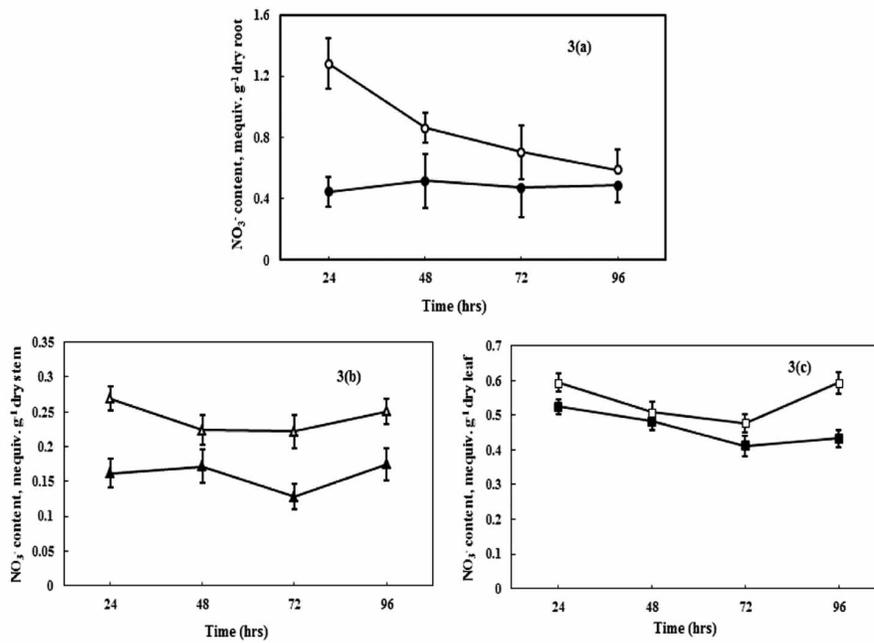


Fig. 3. Effects of Phosphorus deficiency on the accumulation of NO_3^- in (a) root, (b) stem and (c) leaf of mungbean seedlings. Otherwise as Fig. 1.

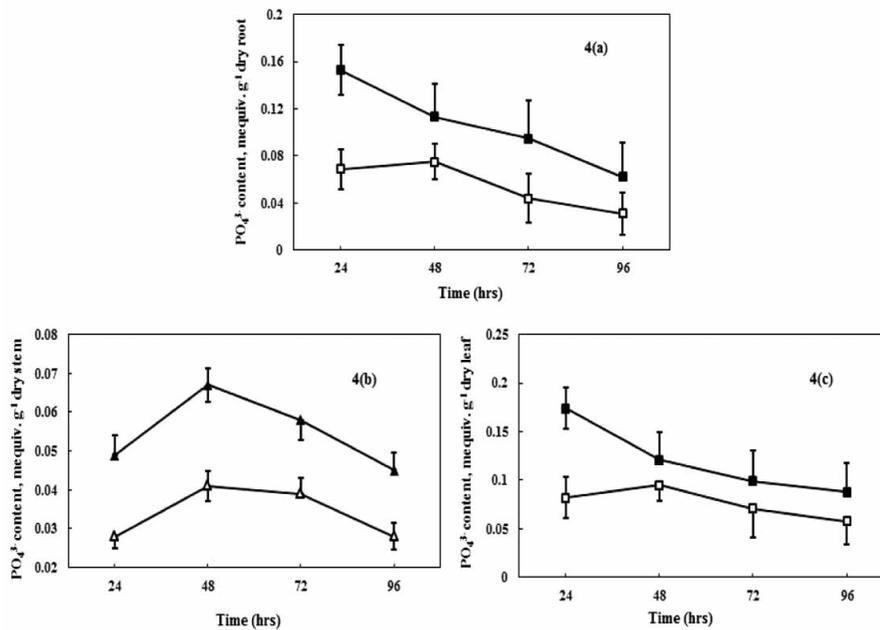
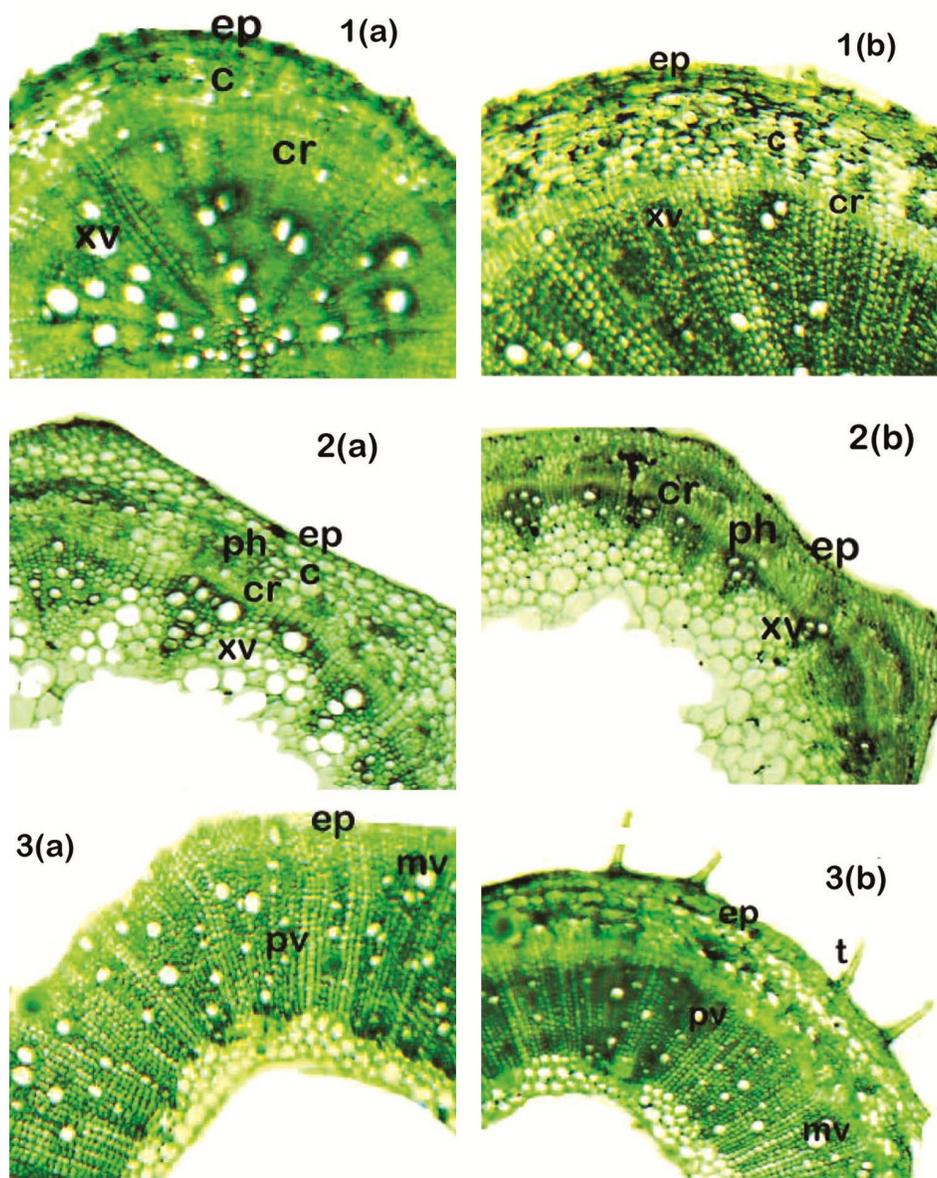


Fig. 4. Effects of Phosphorus deficiency on the accumulation of PO_4^{3-} in (a) root, (b) stem and (c) leaf of mungbean seedlings. Otherwise as Fig. 1.

PO_4^{3-} accumulation decreased by 52.87% at 24 hrs and similar inhibition was observed up to 96 hrs of treatment (Fig. 4a, b and c). Andreeva *et al.* (1992) observed that P deprivation decreased phosphate accumulation in mustard.



Plates 1-3: 1. Transverse section of the root of mungbean ($\times 10$) (a) control (+P) and (b) phosphorus deficient plant (-P) showing epidermis (ep), cortex (c), cambium ring (cr) and xylem vessel (xv). Bar = 100 μm . 2. Transverse section of the primary growth of the stem of mungbean ($\times 10$) (a) control (+P) and (b) phosphorus deficient plant (-P) showing epidermis (ep), cortex (c), phloem (ph), cambium ring (cr) xylem vessel (xv). Bar = 100 μm . 3. Transverse section of the secondary growth of the stem of mungbean ($\times 10$) (a) control (+P) and (b) phosphorus deficient plant (-P) showing epidermis (ep), trichome (t), metaxylem vessel (mv) and protoxylem vessel (pv). Bar = 100 μm .

Under phosphorus deficient condition, root diameter was decreased compared to control. Epidermis was uniseriate and composed of parenchyma cells in both control and phosphorus deficient plant. The parenchyma cells of cortex were large, thin walled and irregular in size both in control and deficient plant. More cortical layer was found in case of P-deficient plant (Plate 1). On the other hand, Sarker and Karmoker (2015) found that phosphorus deficiency-treatment reduced the cortical layer in the root of lentil. Vascular system consists of phloem, xylem and non-lignified pith. The plant showed a smaller root diameter under deficient plant as compared to control. Drastic change in vascular area was noticed in P-deficient root. The number and size of the xylem vessels of phosphorus deficient plant were smaller than those of control plant. Vessel cavity was decreased in P-deficient root compared to control. Phloem tissues were smaller in P-deficient plant. Similar changes in xylem vessels and phloem tissues of lentil root were caused by phosphorus deficiency (Liu *et al.* 2004).

In the stem (internode) of P-deficit mungbean plant, few anatomical differences were observed in relation to the control plants (Plate 2). Transverse section of stem of mungbean grown in +P and -P condition showed monoseriate epidermis. In case of P-deficient plant, vascular bundles were smaller in size. The number and size of the metaxylem vessels of phosphorus deficient plant were reduced than those of the control plant. After secondary growth, the diameter of the cavity of xylem vessels becomes smaller under phosphorus deficient plant as compared to control. In phosphorus deficient plant, phloem elements were decreased in size than that of control plant. Similar effect was also found in wheat (Mehraz *et al.* 2017). Trichome is a hairlike or bristlelike, non-glandular outgrowth from the epidermis of the stem. A number of trichomes were found in the stem (secondary growth) of phosphorus deficient plant (Plate 3).

Phosphorus deficiency in mungbean plant induced decrease in number and diameter of xylem vessels would decrease the translocation of ions from the root to the shoot and thus adversely might affect the distribution of ions in different parts of the plant.

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