

## Effect of arsenate on phosphorus accumulation in rice under simulated condition

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### ABSTRACT

During the study of arsenic (As) accumulation in plant parts, a pot experiment in simulated As condition with different doses of arsenate ( $As^5$ ) viz., 20, 30, 50 ppm and control with four popular rice varieties viz. Triguna, IR 36, PNR 519 and IET 4786 was conducted. It was observed that availability of phosphorus (P) concentration in soil was increased after irrigation with  $As^5$ . Phosphorus accumulation increased in root and husk with increase of  $As^5$  in all the cultivars. P accumulated in increased level in shoot of all cultivars up to 30 ppm of  $As^5$  except in IET 4786 where it increased up to 50 ppm. P accumulation was also increased in seeds of Triguna and IR 36 with the increase of  $As^5$  up to 50 ppm but it had shown reducing effect on P accumulation above 20 ppm in IET 4786 and above 30 ppm in PNR 519.

**Key words:** Arsenate, arsenic, phosphorus, rice cultivars

Inorganic arsenic ( $As_i$ ) is a class I carcinogen (Anon, 2004). This is widespread chronic  $As_i$  poisoning in regions of Asia, South America and elsewhere, due to the consumption of drinking water with geogenically elevated  $As_i$ , with the situation at its worst in the densely populated floodplains and deltas of south and southeast Asia (Brammer and Ravenscroft, 2009; Nordstrom, 2002). Now along with drinking water, plant-based food is also an important source of  $As_i$  contamination.

Consequently rice is a major crop being cultivated in the areas where severe As contamination exists including Bangladesh, India, Taiwan and China (Williams *et al.*, 2005). Rice has been reported to accumulate up to 1.8 mg kg<sup>-1</sup> As in grains and up to 92 mg kg<sup>-1</sup> in straw (Abedin *et al.*, 2002). The total As (mg kg<sup>-1</sup> dw) concentration in rice varies from 0.005 to 0.710 in different varieties and it also differs from one geographical region to other e.g. <0.01-2.05 for Bangladesh, 0.31-0.76 for China, 0.03-0.44 for India and 0.11-0.66 for USA (Zavala and Duxbury, 2008). Arsenic contamination of rice is therefore a newly uncovered disaster on a massive scale. The physical and chemical techniques available for remediation of As has not shown promise to deal with this huge problem (Mondal *et al.*, 2006).

Development of arsenic tolerant rice (Safe grain Arsenic levels for population) through breeding and molecular approaches is an urgent necessity for improving the safe crop productivity in developing countries, particularly in India (Tripathi *et al.*, 2007; Adhikari *et al.*, 2009.).

### MATERIALS AND METHODS

A pot experiment in simulated As condition was conducted in the net house of Rice Research Station, Chinsurah, West Bengal during *boro* 2007-08. Four popular HYVs of rice viz. Triguna, IR 36, PNR 519 and IET 4786 (Satabdi) were selected for

the experiment. Grains were allowed to germinate after surface sterilization (by 0.1% HgCl<sub>2</sub> for 1 min). Transplanting was done with the seedlings of three week. Three seedlings (1 seedlings /hill) of each cultivar were planted at three different places of one pot (14" earthen pot) and the pots were placed into a net house under natural light and humid conditions. Pots were watered daily with deionized water to maintain water logging condition. During tillering, the plants were irrigated with different arsenic concentrations (0, 20, 30 and 50 ppm) and for this Na<sub>2</sub>HAsO<sub>4</sub> were used. Two more irrigations of arsenic were given at pre-flowering and post-flowering stages.

Plants were uprooted carefully and washed thoroughly and brought to the laboratory for analysis. In the laboratory plants were separated into root, shoot, husk and grains. After separation, roots were washed with Milli-Q water. Washed rice roots (1g) were treated with dithionate citrate bicarbonate (DCB) solution (Taylor and Crowder, 1983) to know the level of mineral nutrients adsorbed on the plaque and their relation with As sequestration. pH and EC of soil were measured by ion meter (Orion, USA), while water holding capacity was measured by hydrometry.

P level in rice plant parts and soil including DCB solution, was determined by colorimetric method (Jackson, 1973). As was quantified with the help of inductively coupled plasma mass spectrometer (ICP-MS, Agilent 7500ce) coupled with high performance liquid chromatography (HPLC) and procedure of analysis was performed by following the protocol of Abedin *et al.*, 2002a).

All the experiment was conducted following a randomized block design. Two ways analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) was performed to determine the significant difference between treatments and genotypes.

## RESULTS AND DISCUSSION

The level of P in Fe-plaque increased by increasing the As<sup>V</sup> supply in soil, upto 30 ppm, but at higher As<sup>V</sup> dose (50 ppm) a slight reduction in P was observed in IR 36 and IET 4786 (Fig. 1A). Table 1 showed physico-chemical properties and P and As composition in control pot soil and after supply of different As concentrations. Fe-plaque is commonly formed on the rice roots due to release of oxygen and oxidants into rhizosphere (Liu *et al.*, 2006) and thus differential ability of rice genotypes in terms of oxygen evolution from roots leads to variable Fe-plaque-forming ability and subsequently, variable tendency to accumulate metals and metalloids (Dwivedi *et al.*, 2010). But accumulation of P in the root of IET 4786 (Shatabdi) was exceptionally high at 50 ppm arsenic concentration in compare to the root of other cultivars at the same level. High concentration As and low concentration of P in rice roots indicate that As can competitively inhibit P uptake by roots (Zhang and Duan, 2008) owing to the fact that As is a phosphate analogue and thus both compete for the same transporters (Meharg and Macnair, 1992). The maximum P accumulation (mg kg<sup>-1</sup>) in shoot was found in variety Triguna (388.01) followed by IET 4786 (316.58) and PNR-519 (307.79) and least in IR-36 (305.68). The P content in the shoot increases with the increasing As upto 30 ppm, but increasing trend of P accumulation was observed upto 50 ppm in IET 4786 (Fig. 1C). Zhang and Duan (2008) also reported that shoot P concentration of various tested genotypes decreased due to increased concentration of As. The content of

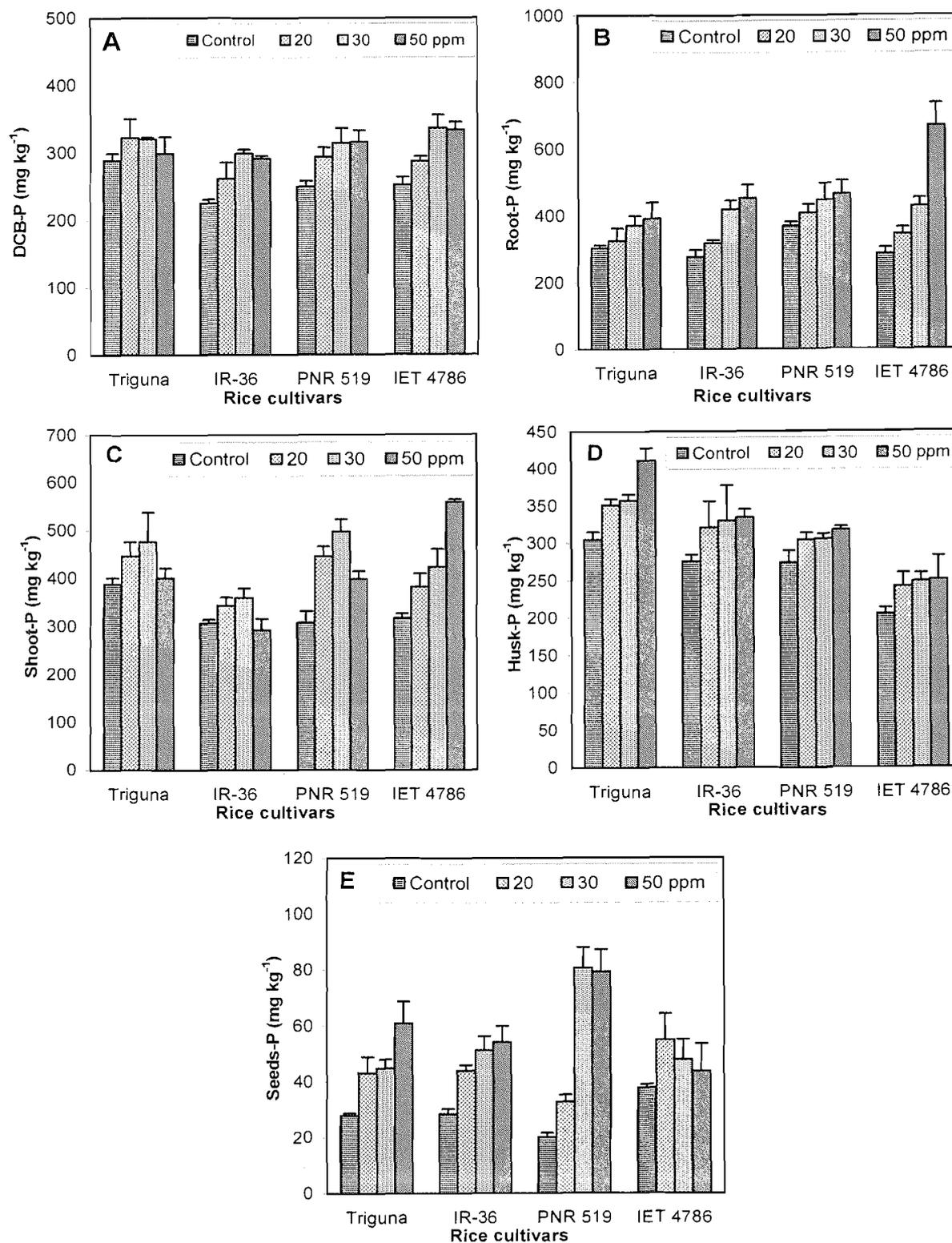
P in husk increased upto 50 ppm in all the varieties and maximum increase was found in Triguna followed by IR-36, PNR-519 and IET-4786 (Fig. 1D). Maximum P accumulation (mg kg<sup>-1</sup>) in seed was found in IET 4786 (37.86) under control conditions and was increased upto 20 ppm; there after it was in decreasing trend. P accumulation was maximum (80.65 mg kg<sup>-1</sup>) in the seed of PNR 519 at 30 ppm As<sup>V</sup>. Increasing trend of P accumulation (mg kg<sup>-1</sup>) in seed was found in Triguna and IR 36 upto 50 ppm As<sup>V</sup> supply. In case of IET 4786, P accumulation was in decreasing rate above 20 ppm As<sup>V</sup> while it was slightly declined above 30 ppm As<sup>V</sup> in case of PNR 519 (Fig. 1E). It was clearly observed from the result that P accumulated at higher amount in root and shoot but at lower amount in husk and seed of all the four cultivars. Increasing or decreasing rate of P accumulation is not in similar order in all the cultivars. In IET 4786, P accumulation was much higher in root and shoots but very low in seed as compare to other cultivars. Thus accumulation of P at different parts of rice plant not only depend on the genotypic differences of the cultivars but also the genetic architecture of the individual cultivar may have some partitioning effect in uptake and transport of P in different parts of the plant along with the supply of irrigation water with As<sup>V</sup>. Zhang and Duan (2008) found significant difference in As uptake and translocation between rice genotype. Rai *et al.* (2011) reported that IET-4786 is very sensitive to arsenic stress due to reduction of both sulphate assimilation pathway and antioxidant defence enzymes in As-detoxification. However Triguna and IR-36 showed considerable detoxification mechanism due to up-regulation of several of these genes during arsenic stress.

**Table 1:** Physico-chemical properties and P and As composition in control pot soil and after supply of different As concentrations

Parameters	Control	As (20 ppm)	As (30 ppm)	As (50 ppm)
pH	7.60 ± 0.32	7.40 ± 0.76	7.30 ± 0.54	7.00 ± 0.44
Electrical conductivity (EC)	176.70 ± 5.66	275.30 ± 8.21	283.00 ± 8.88	332.30 ± 9.21
Total organic carbon (%)	2.21 ± 0.05	2.46 ± 0.04	2.23 ± 0.04	2.30 ± 0.02
Water holding capacity (%)	71.98 ± 3.50	74.79 ± 4.90	75.62 ± 4.10	76.54 ± 5.10
Bulk density (g cm <sup>-3</sup> )	1.26 ± 0.04	1.20 ± 0.04	1.21 ± 0.03	1.18 ± 0.05
Particle density (g cm <sup>-3</sup> )	1.68 ± 0.01	1.72 ± 0.03	1.81 ± 0.02	1.79 ± 0.04
Available P	447.42 ± 11.30	725.69 ± 17.00	605.56 ± 27.20	525.47 ± 16.70
Fe	76146 ± 336.30	75436 ± 300.90	76429 ± 26	73214 ± 299.50
As	5.43 ± 0.23	24.0 ± 1.67	26.27 ± 1.210	31.17 ± 2.81

All the values are mean of triplicates ±S.D. ANOVA significant at  $p \leq 0.01$ .

**Fig. 1 (A-E):** Effect of different concentration of arsenate on phosphorus accumulation in different parts of rice cultivars. A- P in DCB; B- Root P; C- Shoot P; D- Husk P; E- Seed P. All values are the mean of replicates + SD. ANOVA significant at  $p < 0.01$ .



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