



RESEARCH ARTICLE

ACCUMULATION AND TOXICITY IN DIFFERENT PARTS OF WHEAT (*TRITICUM AESTIVUM*) GROWN UNDER ARSENIC STRESS

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ABSTRACT

Arsenic is a toxic heavy metal that enters the environment through various natural and anthropogenic sources and affected various metabolic pathways of plants and animals. Wheat and other plants grown under Arsenic condition accumulate it and decreases growth and productivity. Its toxicity may result from disturbance in plant metabolic activity and disturbance in the uptake and transport of mineral nutrients. Some nutrients and arsenic compete for the same transporters and it accumulate in plant parts to replace nutrient. Arsenic accumulation was measured in plant tissues in different parts at maturity. Experiment was conducted on two varieties of wheat (C-306, WH-711) on three concentration of Arsenic 50, 100 and 150 mg/kg of soil. Experiment showed that higher concentration of arsenic significantly ($p < 0.05$) uptake among both wheat varieties. Arsenic concentrations in plant tissues were as follows: roots > stems > grains. In the arsenic treatments, accumulation in roots was about 3-4 times higher than those in grains. Arsenic enters the plant system, may enter the food chain and cause harm to humans and animals. By observing these parameters of the two varieties of wheat WH-711 is more sensitive and accumulates more arsenic than C-306.

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INTRODUCTION

The increased dependence of agriculture on chemical fertilizers and sewage wastewater irrigation and rapid industrialization has added toxic metals to agricultural soils causing harmful effects on soil-plant environment system. Arsenic (As) is among the metal contamination and is consider as a major environmental concern to the agricultural system (Seth *et al* 2007). Arsenic has been major role in the top toxins. Arsenic concentrations of uncontaminated soils are usually below 0.5 mg·kg⁻¹, but can reach up to high concentration depending on the soil materials. Although Arsenic is a non-essential element for crop plants, it is easily taken up by plants growing on As-contaminated soils, entering food chain and causing damage to plant and human being. The accumulation of Arsenic in plants may cause several physiological and biochemical alterations (Sharma *et al.*, 2007). Arsenic accumulation affect mineral nutrients uptake specially Phosphate and inhibits stomatal opening and closing, changes the internal metabolism, and decrease the crop production. The accumulation of Arsenic in different plant parts plays crucial

role in toxicity of As in plants. The amount of As that accumulates in plant is limited by several factors including 1) As bioavailability within the soil; 2) Rates of As transport into roots 3) The proportion of arsenic fixed within roots as a Arsenic-phytochelatin complex be an important factor influencing the total uptake of As in wheat (Mousavi *et al.*, 2013). The interactions between heavy metals and the properties of soil play an important impact on the environment through their decreasing effect on the availability of heavy metals, thus favorably affecting the environment. High levels of arsenic in soils have been phytotoxic in plants: decreases in plant growth and fruit yields; discolored and stunted roots; withered and yellow leaves (Machlis, 1941); reductions in chlorophyll and protein contents, and in photosynthetic capacity (Marin *et al.*, 1993).

MATERIALS AND METHODS

Plant material and treatments

The plant material which was included in our study was two different wheat varieties (WH-306, WH-711). The seeds were surface sterilized with dilute solution of sodium hypo chlorite to prevent any fungal contamination and then rinsed three

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times with distilled water. The seeds were sown in earthen pots containing equal quantities (4kg) of washed and acid treated loamy sand soil. Metal treatments of Arsenic solutions were prepared with concentrations of 50, 100 and 150 mg/kg of soil. Two identical sets were maintained during the whole experiment. The samples were taken from fully matured stage for physiological analysis. Arsenic determination -Two different species of wheat were collected from experimental pots. Its surface contamination was wiped with tissue paper and left for air drying for 3-4 days the dried sample were powdered in mortar. 10 ml of Nitric acid was added to 2 gm of accurate weight dried plant sample in a 100 ml beaker. The sample was placed on a hot plate and heated at 95°C for 15 minutes. The digest was cooled and add 5 ml Con. Nitric acid and heated for additional 30 minutes at 95°C. The last step was repeated and the solution was reduced to about 5 ml without boiling the sample was cool again and 2 ml of water and 3 ml of 30% hydrogen peroxide was added. With covered beaker, the sample was heated gently to start the peroxide reaction if effervesce becomes excessively vigor's sample was removed from hot plate and 30% hydrogen peroxide was added in 1 ml increments, followed by gentle heating until the effervesces was subsides. 5 ml of concentrated hydrochloric acid and 10 ml H₂O was added and the sample was heated for additional 15 minute without boiling the sample was cool and filtered through Whatman filter 42 and diluted to 60 ml with water. AAS (Atomic Absorption spectrophotometer) was used for determination of Arsenic were carried out in air flame with help of hydride generator.

RESULTS

Arsenic accumulation in plants

Bioaccumulation is an inherent property of plants. On one hand it allows purification of surrounding environment; on another hand, when accumulated in edible plants, it leads to inclusion of the accumulated substance in food chain. To estimate the arsenic accumulation in the wheat plant, we grown the two varieties viz., C-306 and WH-711 in presence of different levels of arsenic and accumulated arsenic in different parts was estimated using atomic absorption spectroscopy as described in the materials and methods section.

Arsenic accumulation in roots

When the two varieties were grown in presence of different concentrations of arsenic, the bioaccumulations were observed in the roots. In variety C306, the arsenic was accumulated to mean 6.13 ± 0.2 mg (HSD Tuskey; p value < 0.01), 10.77 ± 0.25 mg (HSD Tuskey; p value < 0.01) and 16.43 ± 0.55 mg (HSD Tuskey; p value < 0.01) as compared with control having 0 mg of arsenic (Among the group- ANOVA, p value < 0.0001). Whereas, in variety WH-711 the bioaccumulation was found to reach to 7.5 ± 0.4 mg (HSD Tuskey; p value < 0.01), 13.2 ± 0.4 (HSD Tuskey; p value < 0.01) and 19.13 ± 0.31 mg (HSD Tuskey; p value < 0.01) when grown in presence of 50, 100 and 150 mg of arsenic per kg soil in the pot respectively (Among the group- ANOVA, p value < 0.0001).



Figure 1. 2. Show the growth of Wheat(C-306 &WH-711) under different concentration of Arsenic

Table 1. Shows the Accumulation of Arsenic in roots of wheat

Metal Conc./ kg of Soil	C-306(mg/kg.fr.wt)		WH-711(mg/kg.fr.wt)		T-Test p-value
	MEAN	SD	MEAN	SD	
Control	0	0	0	0	0
AS50	6.2	6.13 ± 0.208	7.5	7.50 ± 0.4	0.0038
	5.9		7.1		
	6.3		7.9		
AS 100	10.5	10.76 ± 0.251	13.5	13.2 ± 0.36	0.0072
	11		13.3		
	10.8		12.8		
AS150	16.4	16.43 ± 0.550	19.4	19.13 ± 0.3	0.0042
	15.9		19.2		
	17		18.8		

Variety C306 was found less prone to bioaccumulation in roots since variety WH-711 showed significantly higher accumulation than corresponding amount of arsenic supplied to C306 (T test, p value < 0.01).

Tuskey; p value < 0.01) of arsenic in C-306 (Among the group ANOVA, p value < 0.001); and nearly 2.08 ± 0.07 (HSD Tuskey; p value < 0.01), 3.6 ± 0.26 (HSD Tuskey; p value < 0.01) and 6.23 ± 0.06 mg (HSD Tuskey; p value < 0.01) in

Table 2. Shows the Accumulation of Arsenic in stem of wheat

Metal Conc./ kg of Soil	C-306 (mg/kg.fr.wt)		WH-711(mg/kg.fr.wt)		T-Test p-value
	MEAN \pm SD		MEAN \pm SD		
Control	0	0	0	0	0
	0		0		
	0		0		
AS50	4.01	4.033 \pm 0.04	4.34	4.20 \pm 0.19	0.12
	4.08		4.29		
	4.01		3.98		
AS 100	7.2	7.133 \pm 0.208	8.8	8.8 \pm 0.1	0.0008
	6.9		8.7		
	7.3		8.9		
AS150	10.4	10.66 \pm 0.25	13.5	13.23 \pm 0.378	0.009
	10.9		12.8		
	10.7		13.4		

Table 3. Shows the Accumulation of Arsenic in grains of wheat

Metal Conc./ kg of Soil	C-306 (mg/kg.fr.wt)		WH-711(mg/kg.fr.wt)		T-Test p-value
	MEAN \pm SD		MEAN \pm SD		
Control	0	0	0	0	0
	0		0		
	0		0		
AS50	1.88	1.92 \pm 0.060	2.12	2.07 \pm 0.066	0.083
	1.89		2.11		
	1.99		2		
AS 100	3.2	3.266 \pm 0.057	3.8	3.60 \pm 0.264	0.099
	3.3		3.3		
	3.3		3.7		
AS150	5.2	5.066 \pm 0.152	6.2	6.233 \pm 0.0577	0.0028
	5.1		6.3		
	4.9		6.2		

Arsenic accumulation in stem

Arsenic accumulation in stem was found proportional to the increasing arsenic concentration in the pot. In presence of 50 mg, 100 mg and 15 mg of arsenic per kg soil in the pot mean 4.03 ± 0.04 mg (HSD Tuskey; p value < 0.01), 7.13 ± 0.21 mg (HSD Tuskey; p value < 0.01) and 10.67 ± 0.25 mg (HSD Tuskey; p value < 0.01) of arsenic respectively was accumulated in C-306 variety (Among the group ANOVA, p value < 0.001), whereas, 4.2 ± 0.19 mg (HSD Tuskey; p value < 0.01); 8.8 ± 0.1 mg (HSD Tuskey; p value < 0.01) and 13.23 ± 0.38 mg (HSD Tuskey; p value < 0.01) was accumulated in WH-711 variety (Among the group ANOVA p value, 0.001). Comparison of the two varieties revealed higher accumulation in WH-711 as compared with the C-307 variety. The difference was significantly higher (T test p value < 0.01) when the plants were grown in presence of 100 and 150 mg of arsenic per kilogram of soil in the, pot however the difference was not significant at 50 mg/Kg soil arsenic concentration.

Arsenic accumulation in grains

Arsenic accumulation in grains was found to significantly increase (Among the group ANOVA, p value < 0.0001) with the increasing concentration of the supplied arsenic with average of 1.92 ± 0.06 mg (HSD Tuskey; p value < 0.01), 3.27 ± 0.6 mg (HSD Tuskey; p value < 0.01) and 5 ± 0.15 mg (HSD

variety WH-711 in 50, 75 and 150 mg/Kg of arsenic supplied pot respectively. The difference in bioaccumulation of arsenic in the two strains was again higher in WH-711 as compared with the C306, however the difference was not that big and was found significant only when pot was supplied with 150 mg (T test, p value < 0.005), but not at 50 and 100 mg/ Kg (Figure 1).

Conclusion

The extent of plant injury by elevated concentration is specific and strongly depends on the environmental conditions and on the availability of heavy metals. In the present study we took the two varieties of wheat C-306 and WH-711. These two varieties were grown under Arsenic stress. The metals affect various parameters of wheat at different concentrations. The inhibitory effect of Arsenic on seed germination and seedling growth showed significant linear relationships. It represented that Arsenic is more toxic even at lower concentration. By observing these different parameters of the two varieties of wheat we concluded that the variety WH-711 is more sensitive to heavy metals than C-306. The three arsenic treatments used in this study represent contamination levels in India. The range of As contamination in soils of central India (i.e., Ambargarh Chauki, Chhattisgarh) is 9-105 mg/kg soil. Arsenic concentrations in agricultural soils of As-affected areas in Bangladesh range from 20 to 90 mg/kg soil. Our study showed

that As levels of 150 mg/kg & 100 mg/kg soil reduced wheat seed germination rates and inhibited seedling growth. The two varieties tested in this study differed greatly in their response to As addition in soil, they followed the same pattern: when As was added, a negative response by plant part biomass and yield components, and a positive response by plant parts at wheat maturity were obtained. Damaging effects of As on plants might also be due to effects on phosphate uptake and utilization. Phosphate level is known to control plant growth and development, and arsenate is a phosphate analogue. Irrespective of the As treatment, roots contained higher concentrations of As than stem. Arsenic concentration order, however, was roots > stems > >grains in two wheat varieties. Flour is the main edible portion in wheat grains. Arsenic content in flour is critical for human health.

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