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Arsenic Accumulation in Rice (*Oryza sativa L.*) Genotype of India

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Abstract

Rice is prone to arsenic accumulation due to its cultivation in water logging condition favouring arsenic mobility. Arsenic in rice depends on its availability due to its irrigation with arsenic contaminated water. Therefore it is necessary to resolving this human health risk. Screening of rice genotypes with low arsenic accumulation is considered as the most reasonable and effective approach. The present study was done on arsenic accumulation in common rice genotypes grown in hydroponic medium. All rice seeds were germinated and grown in presence of two concentration of arsenic 50µM and 250µM with control. The results revealed that arsenic concentration in all the genotype increased significantly (p<0.05) with increase in arsenic treatment. According to results Ndr359 genotype accumulated highest concentration of arsenic and swarnsub1 accumulated low arsenic. All the twenty rice genotypes were ranked with respect to their As tolerance index: Ndr3>Sarj>Swarn>Brh5>Laxm>Hkr4>Bg90>Jkrh>Pr11>Nk33>Mtu7>Suru>Ndr8>Pant>Sarb>Ndr9>Bpt5>Tcs5>Nare>Pb01. The results revealed that LARG is the most appropriate to grow in arsenic contaminated region of India.

Keywords: Arsenic, rice genotype, translocation factor, screening, tolerance index.

Introduction

Soil contaminated by heavy metal pollution received attention because most of the environmental pollutant comes from industrial and agricultural source affects the soil productivity. Arsenic is hazardous environmental pollutant and toxic metalloid because of its chronic and epidemic impact on human health. It comes from both natural and anthropogenic sources such as mining and agricultural activities [1,2] and enters into the terrestrial and aquatic environments. Incidence of arsenic occurrence in the ground water has been reported in many part of the world. About 20 countries round the world have been reported to contain as naturally in their aquifer rocks. These countries include Thailand[3,4], Greece, Hungry, Inner Mongolia, USA, Argentina, Ghana, Chile and Mexico[5,6] are the highly contaminated region in all over the world. In Asian region Bangladesh and West Bengal in India are extremely contaminated region of Asian continent where ground water rich in arsenic[7-13]. Using this arsenic contaminated water for irrigation purpose, arsenic gradually accumulated into the crops and vegetables and entered into the food chain causing human health risk. Rice (*Oryza sativa L.*) is one of the predominant staple crops all over the world, especially in Southeast Asia. It has been consumed as a principle food source of 3 billion people of the world[14]. The amount of arsenic present in rice is comparatively higher than present in other plants[15]. Rice as compared to other crops has potential to accumulate arsenic because of its cultivation in water logging condition favouring arsenic mobility. Rice grain has been reported to accumulate arsenic upto 2.0 mg/kg in grain[16,17] and up to 92 mg kg^{-1} in straw. According to WHO recommendation the permissible limit of As in rice is (1.0 mg/kg)[18]. Plant species and even genotypes differ significantly in their ability to uptake, transport and accumulate arsenic into the biomass. The objective of the present study was to screen rice genotypes with low arsenic accumulation is the most feasible and effective approach.

Material and Methods

Experiment was done in controlled environment. The day/night regime was 14/10h light/dark period (260-350mmol m-2.s-1) with an average temperature of 25˚C during day and 20˚C during the night and relative humidity was 50-70%. All the germinated rice seedlings were transferred into the plastic trays containing nutrient solution having (50µM and 250 µM) of arsenic doses. Nutrient solution was changed twice a week and pH of the solution was adjusted to 5.5 using 0.1 M KOH and HCl. After 15 days of treatment all plants were harvested.

Quantification of Arsenic Concentration: All plant samples were separated into root and shoot parts and kept for drying at 65˚C in oven for two days. Dried plant sample was mixed with nitric acid and perchloric acid (5:1 v/v) and digested in microwave digester (Speed wave digester BERGHOF speed wave Digester unit MWS-3+). The digested samples were filtered with Whatman-42 filter paper and diluted with distilled water and arsenic was estimated by inductively coupled plasma mass spectrometry (ICP-MS) Agilent Technologies 7500 Series.

Quality control and quality assurance: The standard reference material of arsenic (consisting of 998±4 mgl^{-1}As-NIST and BAM-CRM traceable; EMerck, Germany) was used for each analytical batch. Analytical data quality was ensured with repeated analysis of quality control samples (n=3) and the results were within (±2.82 mgl^{-1}) limit of the certified values. Standard AA03N-3 (Accustandard, USA) was used as a matrix reference material which was spiked with known concentration (0-50 μgl^{-1}As) of standard reference material, and the recovery of total As was within 85.3% (±2.8; n=5) to 89.5%.

Statistical analysis: Effect of arsenic on growth parameters root, shoot length, biomass and arsenic uptake data in the tested rice genotypes was analysed by Duncan multiple range test (DMRT) by using SPSS 16 software package.

Results and Discussion

Twenty rice (*Oryza sativa L.*) genotypes treated with two levels of arsenic showed variations in respect to root-shoot length, biomass production and arsenic accumulation. Root, shoot length, plant biomass are valuable parameters in the studies of metal resistance to plant[19,20]. Different parameters were analyzed separately to obtain significant data to process consecutively to make the comparison more reliable. Biomass and root-shoot length in a medium are commonly used for investigating vegetative responses in the studies on the resistance of plants to metals stress[19,20]. In 50µM as treatment Mtu, Bg90 and Ndr359 genotype had longest root length hence these genotypes showed tolerance in presence of arsenic table-1. While Sarbati, Ndr97 and Bpt genotypes had lowest root length hence root growth was vigorously effected in these genotypes while remaining genotype showed intermediate results. Growth of rice root in presence of metal availability has been reported as more sensitive point than chlorophyll assays[21]. Longest shoot length was recorded in swarnsub1 genotype and low was recorded in Bpt genotype in 50µM as treatment. In higher as treatment 250µM roots length ranged from (2.1 to 7.2 cm) and shoot length ranged from (5.9 to 12.8cm). Mtu and Sarbati genotype showed elongation in root and shoot length while inhibition in root shoot length occurred in Pr113 and Bpt genotype table-1. Therefore root in these genotypes exhibited stunted growth after exposure to As. There were no significant variations in the plant dry weight in control treatment, whereas a significant decrease in plant biomass was observed in all the genotypes exposed to as concentrations 50µM and 250µM, compared with the control. Plant biomass was high in Bg90, Mtu and Ndr359 genotype in presence of 50µM arsenic treatment. While Bpt, Brh and Narendra usar had low biomass in 50µM arsenic treatment rest of the genotypes showed mixed results. In 250µM as treatment Ndr359 had highest biomass and Bpt genotype had low biomass. All rice genotypes were categorized decrease in biomass as follow: Ndr359>Bg90>Pr113>Mtu>Hkr>Jkrh>Sarju>Pant>Ndr8>Laxmi>Suruchi>Swarnsub>Tcs>Nk33>Ndr97>Pb1>Sarbati>Brh>Narendra usar>Bpt genotype respectively

Table-1
Effect of two arsenic treatments in root length and shoot length of twenty rice genotypes. Mean values in a column having same letter do not differ significantly at p≤0.05 level by Duncan's multiple range test

Rice Genotype	Root length Control	Root length 50µM As	Root length 250µM As	Shoot length Control	Shoot length 50µM As	Shoot length 250µM As
BRH-5	5.77±0.6[a]	3.93±0.9[b]	2.47±0.5[c]	11.63±1.1[a]	9.77±1.3[ab]	8.03±2.0[a]
JKRH 401	6.13±0.5[a]	3.27±0.5[b]	2.23±0.6[b]	7.83±0.7[ab]	8.80±0.8[a]	7.03±0.8[b]
Suruchi 5629	3.87±0.3[a]	3.53±1.1[a]	3.33±1.4[a]	9.93±0.6[ab]	9.77±1.4[a]	7.30±0.9[b]
Laxmi	4.77±0.6[a]	3.83±0.7[a]	3.53±0.8[a]	8.67±0.7[b]	11.13±0.9[a]	8.67±1.0[b]
MTU7029	8.70±0.6[b]	7.83±0.6[a]	7.20±0.7[a]	10.30±1.7[a]	9.57±2.0[a]	8.77±0.7[a]
BG90	6.67±1.7[b]	5.77±1.1[a]	4.37±1.1[c]	10.10±0.6[a]	9.90±0.9[a]	8.40±0.3[a]
NDR359	6.07±0.7[a]	5.47±0.9[a]	3.77±0.9[b]	8.67±0.8[a]	10.13±1.0[a]	9.90±0.6[a]
SARJU52	4.30±0.7[ab]	4.27±0.4[a]	2.83±0.8[b]	9.23±0.3[a]	8.57±1.1[a]	8.27±0.3[a]
NK33-25	5.47±1.2[a]	3.20±0.5[b]	2.83±0.6[b]	10.03±0.5[a]	8.00±0.8[b]	7.53±0.5[b]
HKR47	5.47±1.3[ab]	4.47±0.3[a]	3.70±0.6[b]	8.47±1.3[a]	7.53±0.4[a]	6.03±0.9[b]
Narendra User	4.10±0.8[a]	4.00±0.3[a]	2.40±1.0[b]	11.83±0.5[b]	10.83±0.7[a]	9.83±1.2[ab]
Swarnsub1	4.07±0.6[a]	2.83±0.6[b]	2.67±0.3[b]	14.60±1.8[b]	12.67±0.3[a]	9.50±1.3[b]
PB1	6.00±1.5[a]	4.20±0.3[a]	3.67±1.9[a]	10.77±1.3[a]	10.50±1.3[a]	9.17±0.5[a]
BPT5204	4.20±0.8[a]	2.73±0.8[b]	2.13±0.5[b]	7.77±1.8[a]	6.97±0.8[a]	5.97±0.7[a]
PR113	3.77±0.2[a]	3.27±1.0[ab]	2.10±0.4[b]	12.37±1.0[a]	9.67±1.0[ab]	8.90±2.3[b]
PANT12	4.13±1.4[a]	3.20±1.1[a]	2.50±0.5[a]	13.90±1.3[a]	12.57±2.7[a]	10.93±0.4[a]
NDR97	3.80±1.1[a]	2.70±0.6[a]	3.23±0.3[a]	9.50±2.6[a]	10.30±0.8[a]	8.50±1.3[a]
NDR8002	4.90±0.3[a]	4.00±0.4[a]	5.13±1.3[a]	13.20±1.7[a]	10.23±1.6[b]	9.67±1.0[b]
Sarbatib Asmati	3.13±1.0[a]	1.90±0.1[a]	3.33±1.0[a]	8.13±1.1[b]	7.27±0.3[b]	12.80±0.8[a]
TCS555	2.5±0.4[a]	3.07±0.5[a]	2.77±1.3[a]	10.60±1.2[a]	10.60±0.9[a]	11.07±1.7a

Table-2
Effect of two arsenic treatments on biomass of twenty rice genotypes. Mean values in a column having same letter do not differ significantly at p≤0.05 level by Duncan's multiple range test

Rice Genotype	Plant biomass Control	Plant biomass 50µM As	Plant biomass 250µM As
BRH-5	16.30±2.9[a]	13.43±0.5[ab]	10.53±1.0[b]
JKRH 401	22.47±1.9[a]	18.60±2.3[b]	14.20±1.7[c]
Suruchi 5629	21.10±2.8[a]	18.63±1.3[a]	12.77±1.0[b]
Laxmi	19.90±1.5[a]	14.83±0.4[b]	13.03±1.7[b]
MTU7029	28.43±1.0[a]	22.10±2.7[b]	17.33±1.4[c]
BG90	29.57±3.2[a]	22.40±1.3[b]	19.00±2.9[b]
NDR359	27.90±2.5[a]	21.20±0.6[b]	21.07±1.3[b]
SARJU52	19.37±1.2[a]	17.57±2.0[a]	13.57±0.2[b]
NK33-25	19.17±0.3[a]	15.57±0.8[b]	11.93±0.4[c]
HKR47	24.70±1.6[a]	17.80±1.6[b]	15.97±2.1[b]
Narendra User	19.70±0.5[a]	14.27±1.5[b]	10.17±2.5[c]
SWARNSUB1	19.30±0.5[a]	16.20±3.0[ab]	12.77±1.2[b]
PB1	25.43±4.8[a]	18.37±1.7[b]	11.83±1.5[c]
BPT5204	17.67±1.1[a]	12.87±1.6[b]	9.97±0.5[c]
PR113	27.53±3.0[a]	19.46±1.9[b]	17.47±1.9[b]
PANT12	24.30±6.0[a]	17.30±2.4[ab]	13.40±0.7[b]
NDR97	21.03±0.9[a]	15.45±3.1[b]	11.87±1.2[b]
NDR8002	23.10±5.0[a]	16.17±2.5[b]	13.27±1.7[b]
Sarbatib Asmati	20.20±0.4[a]	17.13±2.4[a]	11.47±1.9[b]
TCS555	21.97±1.3[a]	15.53±1.6[b]	12.10±0.7[c]

Arsenic concentration in rice genotype and translocation factor: Accumulation of arsenic in rice depends on the genotypes. Concentration of arsenic in all the rice genotype increases significantly ($p<0.05$) with increase in arsenic treatment in nutrient solution. Arsenic accumulation in rice plant has been extensively studied by several authors[22,23]. According to data as concentration in control rice genotypes was below the detection limit. In 50µM as highest concentration of as was recorded in Bg90, Ndr359, Pb1, Laxmi, Jkrh, Mtu7 genotype. While lowest arsenic concentration was recorded in Hkr4, Narendra, Swarnsub1, Nk33, Pant12, Sarbati and the remaining genotype showed intermediate results figure-1. In higher arsenic treatment 250µM significant ($p<0.05$) variation was recorded in each rice genotype. Ndr359 genotype accumulated highest concentration of arsenic and lowest arsenic was recorded in swarnsub1 genotype. Based on present results Ndr359 genotype categorized as high arsenic accumulating rice genotype (HARG) and Swarnsub1 categorized as low arsenic accumulating rice genotype (LARG).Translocation factor was also evaluated to measure the transfer of arsenic from root to shoot part in all the tested rice genotypes. According to results at 50µM As treatment highest TF was recorded in Mtu7 genotype followed by Ndr359, Brh5, Bg90, Suruchi, Laxmi rice genotype. Low TF was recorded in Hkr4 and Nk33 genotype figure-2. At higher arsenic treatment Suruchi was recorded highest TF indicated its high capacity to transfer arsenic from root to shoot. Lowest TF was recorded in Pant>Tcs5>Ndr9>Bpt5>Sarbati genotype figure-2 and in rest of the genotype results was intermediate. Tolerance index (Ti) was estimated to measure the tolerance of tested rice genotype in presence of arsenic [24]. Overall inhibitory effect of arsenic on each genotype was recorded. At 50µM as treatment Ndr8002 and Pr113 had low (Ti) value and Sarju52 and Suruchi was recorded highest (Ti) value. In 250µM as treatment difference in (Ti) value was high, Ndr359 and Sarju52 rice genotype showed highest (Ti) value hence these genotypes tolerate high arsenic compared to other rice genotype. Lowest (Ti) value was recorded in Pb1 and Narendra genotype. All the rice genotype was categorized in decreasing value of tolerance index as follows:Ndr359>Sarj>Swar>Laxmi>Hkr4>Brh5>Bg90>Pr11>Jkrh>Nk33>Mtu7>Suru>Ndr8>Sarb>Ndr9>Bpt5>Pant>Tcs5>Nare>Pb1 respectively figure-2.

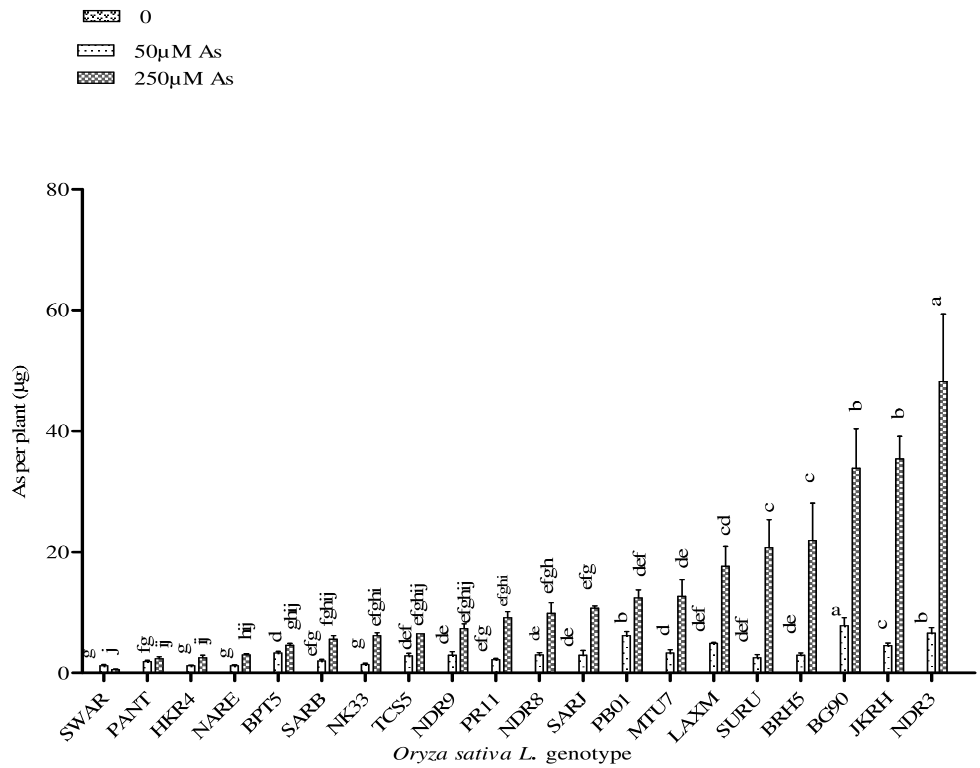


Figure-1
Shows arsenic concentration in twenty rice genotypes to the applied two arsenic treatments 50 and 250 µM

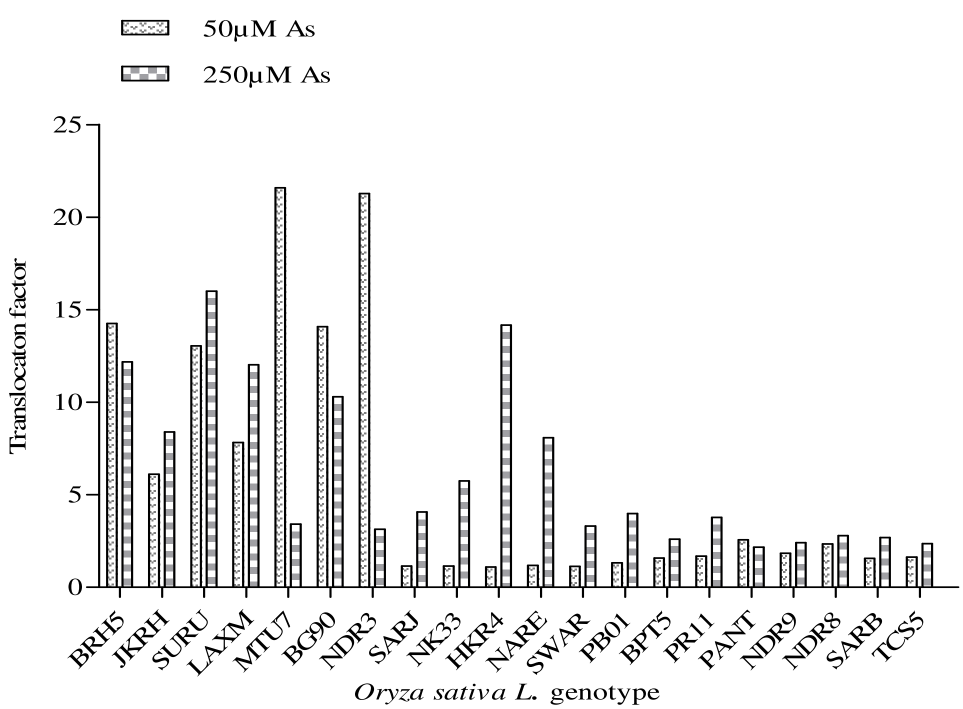
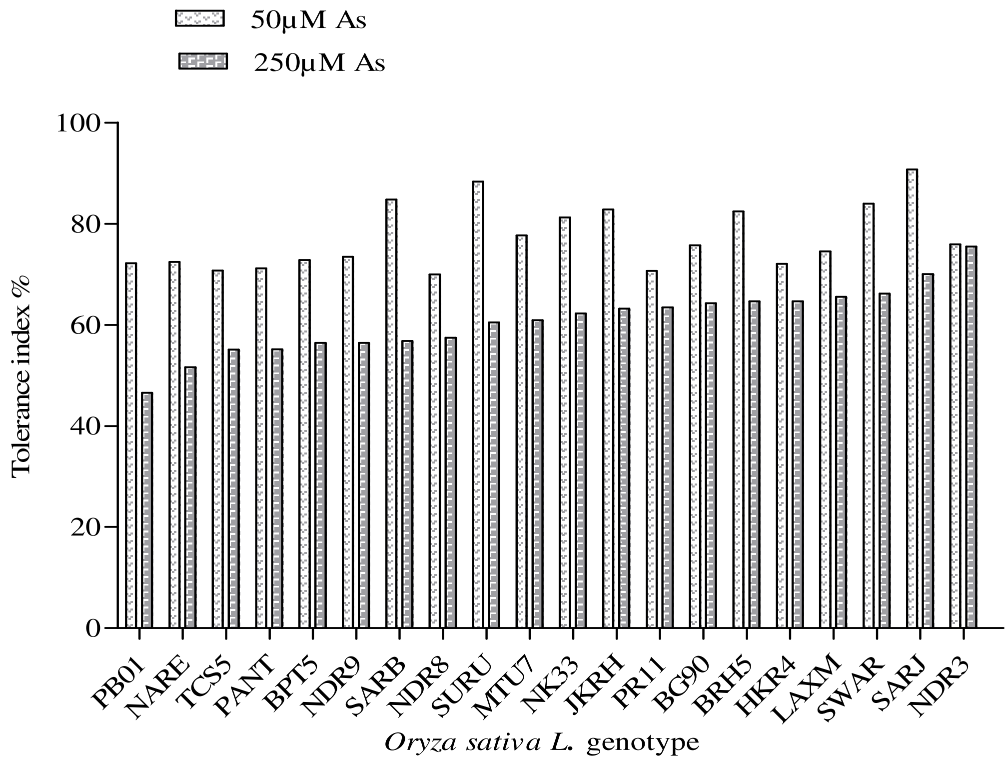


Figure-2
Shows translocation factor (TF) of twenty rice genotype to the applied two arsenic treatments 50 and 250µM



Figure–3
Shows tolerance index (Ti%) of twenty rice genotype to the applied two arsenic treatments 50 and 250µM

Conclusion

In the present study screening test were done and found difference in as accumulation patterns in all the rice genotypes. Large variation occurs in tested genotypes and they behave differently in two arsenic treatment 50 and 250 µM. The results indicates that Ndr359, Jkrh, and Bg90 genotype can both tolerate and accumulated high As so categorized as high arsenic accumulating rice genotype (HARG). While Swarnsub1, Pant12, Hkr47 rice genotype accumulated low arsenic categorized as low arsenic accumulating rice genotype (LARG). Hence (LARG) genotypes are the most appropriate for growth on arsenic contaminated area with economical produce. (LARG) can be used in production in arsenic contaminated region of India.

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Reference

1. Wang Z.G., He H.Y., Yan Y.L., Wu C.Y., Yang Y. and Gao X.Y., Arsenic exposure of residents in areas near Shimen arsenic mine., *J. Environ. Health,* 16, 4–6 **(1999)**
2. Xie Z.M., Liao M. and Huang C.Y., Effects of arsenic pollution on plants and human health and counter measures, *Guangdong Trace Element Science*, **4**, 17–21 **(1997)**
3. Visoottiviseth P., Francesconi K. and Sridokchan W. The potential of Thai indigenous plant species for the phytoremediation of arsenic contaminated land, *Environ Poll*, **118**, 453–461**(2002)**
4. O'Neill P. Arsenic., In Alloway, B.J. (Ed.), Heavy Metals in Soils., 105–121 **(1995)**
5. Smedley P.L. and Kinniburgh D.G., A review of the source, behaviour and distribution of arsenic in natural waters, *Appl Geochemis*, **17**, 517–568 **(2002)**
6. Banerjee D.M., Some comments on the source of arsenic in the Bengal Deltaic sediments. In: Bhattacharya, P., Welch, A.H. (Eds.), Arsenic in groundwater of sedimentary aquifers. 31st International geological congress, Rio de Janerio, *Brazil,* 15–17 **(2000)**
7. Chakraborti A.K. and Das D.K., Arsenic pollution and its environmental significance, *Interacad.1*: **J.272**. 262–276 **(1997)**
8. Fazal M.A., Kawachi T. and Ichio E., Validity of the latest research findings on causes of groundwater arsenic contamination in Bangladesh, *Water Internat,* **26(2)**, 380–389 **(2001)**

9. Hopenhayn C., Arsenic in drinking water : Impact on human health, *Elements,* **2,** 103–107 **(2006)**

10. Smith A.H., Lingas E.O. and Rahman M., Contamination of drinking water by arsenic in Bangladesh: A public health emergency, *Bulletin of the World Health Organization,* **78(9),** 1093–1103 **(2000)**

11. Nickson R., McArthur J., Burgess W. and Ahmed K.M., Ravenscroft P, Rahman M. Arsenic poisoning of Bangladesh ground water, *Nature*, 395- 338 **(1998)**

12. Nickson R.T., McArthur J.M., Ravenscroft P., Burgess W.G. and Ahmed K.M., Mechanism of arsenic release to groundwater, Bangladesh and West Bengal, *Applied Geochemistry,* **15**, 403–413 **(2000)**

13. Stone R., Arsenic and paddy rice a neglected cancer risk, *Nature,* **321**, 184–185 **(2008)**

14. D'Ilio S., Alessandrelli M. and Cresti R. et al, Arsenic content of various types of rice as determined by plasma-based techniques, *Microchemi. J.*, **73**, 195–201**(2002)**

15. Islam M.R., Jahiruddin M., Rahman G.K.M.M., Miah M.A.M., Farid A.T.M., Panaullah G.M., Loeppert R.H., Duxbury J.M. and Meisner C.A., Assessment of arsenic in the water-soil-plant systems in gangetic flood plains of Bangladesh, *Asian J of Plant Sci*, **3**, 489-493**(2004)**

16. Meharg A. A., Arsenic in rice - understanding a new disaster for South-East Asia, *Trends in Plant Sci,* **9**, 415–417 **(2004)**

17. Ohno K., Yanase T., Matsuo Y., Kimura T., Rahman M.H., Magara Y. and Matsui Y., Arsenic intake via water and food by a population living in an arsenic-affected area of Bangladesh, *Sci of the Total Envi*, **381**, 68-76 **(2007)**

18. Abedin M.J. and Meharg A.A., Relative toxicity of arsenite and arsenate on germination and early seedling growth of rice (*Oryza sativa* L.), *Plant Soil*, **243**, 57–66 **(2002)**

19. Abedin M.J., Feldman J. and Meharg A.A., Uptake kinetics of arsenic species in rice plants, *Plant Physio*, **128**:1120–1128 **(2002)**

20. Morgan A.J., Evan M., Winters C., Gane M and Davies M.S., Assaying the effect of chemical ameliaorents with earthworms and plants exposed to a heavily polluted metalliferous soil, *Euro J of Soil Biol.,* **38**, 323-327 **(2002)**

21. Marin A.R., Masscheleyn P.H. and Patrick W.H., The influence of chemical form and concentration of arsenic on rice growth and tissue arsenic concentration, *Plant Soil*, **139**, 175–183**(1992)**

22. Marin A. R., Pezeshki S.R., Masschelen P.H. and Choi H.S., Effect of dimethylarsenic acid (DMAA) on growth, tissue arsenic, and photosynthesis of rice plants, *J of Plant Nutri*, **16**, 865–880 **(1993)**

23. Xie Z.M., Liao M., Huang C.Y., Effects of arsenic pollution on plants and human health and counter measures, Guangdong Trace Element Science, **4**, 17–21 **(1997)**

24. Wilkins D.A., The measurement of tolerance to edaphic factors by means of root growth, *New Phyto*., **136**, 481-488 **(1978)**