Research Article **Allied Science**

International Journal of Pharma and Bio Sciences

ISSN 0975-6299

BIOACCUMULATION AND TRANSPORT OF ARSENIC IN DIFFERENT GENOTYPES OF LENTIL (LENS CULINARIS MEDIK.)

DIBYENDU TALUKDAR*

Plant Cell and Stress Biology Lab, Department of Botany, R.P.M. College (University of Calcutta), Uttarpara, Hooghly 712258, West Bengal, India.

ABSTRACT

Eight genotypes of lentil (*Lens culinaris* Medik.) were tested for bioaccumulation and transport of arsenic (As) in a pot experiment under controlled growth condition. Along with control (no As was added), plants were grown for 60 d in pot soil spiked with 100 mg As kg^{-1}, added as Sodium arsenate. Significant ($P < 0.05$) variations among eight genotypes were observed in bioaccumulation and transport of As in different plant parts. In general, roots contained higher amount of As than shoots, resulting in lowering of bioaccumulation factors but rise in the value of bioconcentration factors. The upward transport of As from roots to shoots was evidenced by moderate to high transfer factors and enrichment factors in six of the eight genotypes studied. L 414 and L 830 are relatively safe for edible purposes. The results suggested genotypic differences in As accumulation in lentil crops grown in As-contaminated regions.

KEYWORDS: **Arsenic, Bioaccumulation, Transport, Nutrition, Lens culinaris**

DIBYENDU TALUKDAR
Plant Cell and Stress Biology Lab, Department of Botany, R.P.M. College (University of Calcutta), Uttarpara, Hooghly 712258, West Bengal, India.

****Corresponding author***

INTRODUCTION

Arsenic (As) is a ubiquitous toxic metalloid without known biological functions in higher plants [1]. Being an analogue of phosphate, arsenate is readily taken up by plants through high-affinity phosphate transporters [2]. Major crops such as cereals and legumes grown in As contaminated fields accumulate substantial amounts of As in their edible parts that may pose huge health risks [3, 4]Groundwater contamination by As and its entry into crops through water-soil-plant system has caused great environmental concern because As is a potent mutagen and a powerful carcinogen [5, 6].

Lentil (*Lens culinaris* Medik.) is one of the oldest known pulse crops and among the earliest of humankind's plant domesticates and associated with the start of the 'agricultural revolution' about 10,000 years ago in the Near East [7].Within the cool season legumes, it is the richest in the important amino acids (lysine, arginine, leucine and sulphur containing amino acids) [8]. More than 85% of the annual global production occurs in four specific regions in which eastern half of the Indo-Gangetic plain of south Asia including India, Nepal, and Bangladesh occupies the major (32%) share [9]. However, the declining/unstable trend of its production in many lentil growing countries is a major cause of concern for food and nutritional security throughout the world [10]. One of the major causes of dwindling lentil production is its sensitivity to diverse types of abiotic stresses including cold, drought, heat, salinity, and nutrient toxicity [8, 11, 12, 13]. Although lentil shows higher sensitivity to abiotic stress than broadbean and soybean [12] and large parts of lentil cultivated areas in several countries including Gangetic Bengal delta and Bangladesh are As-contaminated, perusal of literature cites only scanty information regarding response of this legume crop to As [14]. In view of the wide-scale contamination of agricultural fields by As-contaminated irrigated water in India, Bangladesh and other pulse growing countries in South-East Asia [15] and the reported toxicity of As on growth and nutritional quality of many leguminous crops [14, 16] the accumulation of this toxic metalloid in plant parts needs urgent study. No comprehensive reports are available regarding As accumulation and its transport to photosynthetic and edible parts of lentil. Thus, the present investigation was undertaken to analyse the transport and accumulation of As in different parts of eight lentil genotypes grown widely in As-contaminated areas of Gangetic West Bengal.

MATERIALS AND METHODS

(i) Plant material and As treatments

Fresh and healthy seeds of eight varieties of lentil (*Lens culinaris* Medik. cv. IPL 81, IPL 406, L 414, DPL 59, L 830, VLM 1, VLM 4 and a local check) grown widely in Gangetic West Bengal were surface sterilized in 1% mercuric chloride solution for 15 min followed by 70% ethanol wash for 5 min. Seeds were then washed thrice in sterile distilled water, taken in Petri dishes containing sterile half strength of Murashige and Skoog medium and allowed to germinate under 14 h photoperiod at 25 ºC. Germinated seeds were immediately transplanted to earthen pots of equal sizes (30 cm in height and 40 cm in diameter) with six seedlings per pot. The soil was mixed, air-dried, and ground to a particle size of < 2 mm. Eight kg of air-dried soil was put in each pot. Plants were grown in a clean soil (control) and a soil spiked with 100 mg As kg^{-1}, added as sodium arsenate (As, MW 312.01 g/mol; technical grade, purity 98.5 %, Sigma-Aldrich, Bangalore, India). Each treatment had 4 replicates. The pots were arranged in a completely randomized way, and the plants were grown in a controlled environment (Temperature 27 ºC ± 2 ºC, relative humidity 70 % ± 3 %, photoperiod 14/10 h day/night, PPFD-200 µmol m^{-2} s^{-1}). The plants were thinned to 4 per pot three weeks later and to one plant per pot one month later. Sixty days after sowing, the plants were harvested and washed with deionized water. The plants were then separated into shoots and roots.

(ii) Sample treatment and digestion

The plant samples were washed thoroughly in As-free water to remove adhered soils and dusts, then rinsed in de-ionized water, immersed for 10 min in 10×10^{-3}M KH_2PO_4 solution (pH 6.0) to remove external inorganic As from the root surface and blotted dry. The washing of the plant samples were finished as fast as possible to avoid any possible leakage of absorbed As [17]. The leaves/leaflets were hand separated. Roots, stems, pods and seeds were then separated carefully, oven-dried at 105 °C for 24 h and were stored in airtight polyethylene bags at room temperature with proper labelling. Soil and plant (parts) samples were digested separately following heating block digestion procedure [6] with slight modifications. Of the sample, 0.5–1.0 g was taken into clean, dry digestion tubes, and 5 ml of concentrated HNO_3 was added to it. The mixture was allowed to stand overnight under fume hood. In the following day, the digestion tubes were placed on a heating block and heated at 60 °C for 2 h. The tubes were then allowed to cool at room temperature. About 2 ml of concentrated $HClO_4$ was added to the plant samples. For the soil samples, 3 ml of concentrated H_2SO_4 was added in addition to 2 ml of concentrated $HClO_4$. Then, the tubes were heated at 160 °C for about 4–5 h. The heating was stopped when the dense white fume of $HClO_4$ was emitted. The content was then cooled, diluted to 25 ml with de-ionized water, and filtered through Whatman No. 41 filter papers and finally stored in polyethylene bottles. Prior to sample digestion, all glass goods were washed with 2% HNO_3 followed by rinsing with de-ionized water and drying.

(iii) Chemical analysis of As

The total As of samples was analyzed by flow injection hydride generation atomic absorption spectrophotometer (FI-HG-AAS, Perkin Elmer AAnalyst 400, lamp current 400 mA, wavelength 194 nm, with flow rate of 1.2 ml min^{-1} for HCl, of 2 ml min^{-1} for $NaBH_4$, and 130 ml min^{-1} for carrier gas N_2). For each sample of the digested soil and plant parts, four replicates were taken and the mean values were obtained on the basis of calculation of those replicates. All chemicals were of analytical grade, and distilled de-ionized water was used throughout the experiment. Standard Reference Materials (SRM) of tomato leaves (SRM 1573a) and of San Joaquin soil (SRM 2709a) from National Institute of Standards and Technology, USA were analyzed in the same procedure at the start, during and at the end of the measurements as part of the quality assurance/quality control protocol. The values (mg kg^{-1} DW) of As concentration in certified (0.112 ± 0.004 for dried tomato leaves and 17.7 ± 0.8 for San Joaquin soil) and my measured samples (0.117 ± 0.01 for tomato leaves and 16.6 ± 0.11 for soil) were in good correlations ($r = 0.89$ for tomato leaves and $r = 0.82$ for soil, $P < 0.05$, $n = 10$).

(iv)Assessment of bioaccumulation of As

The bioaccumulation factors (BFs) correspond to the concentration of the same metal present in the shoots divided by the concentration (mg kg^{-1} DW) of that metal in the soil. The bioconcentration factor (BCFs) were calculated by dividing root As concentration with soil As concentrations. Transfer factors (TFs) were calculated from the metal concentration of As in aboveground parts divided by metal concentration in roots. Enrichment factors (EFs) were calculated by dividing As concentration in edible part of a plant grown in As-contaminated soil with As concentration in edible part of a plant grown in control soil [18]. In the present study, leaves, stems, and pods together were considered as shoot, while seeds were considered as edible part.

(v) Statistical analysis

The results presented are the mean values ± standard errors of at least four replicates. Multiple comparisons of means were performed by ANOVA (SPSS Inc. v. 10), and the means were separated by Duncan's Multiple Range Test considering significant differences at $P < 0.05$.

RESULTS

Significant ($P < 0.05$) differences were observed between genotypes regarding transport and accumulation of As in different

plant parts of lentil crop. Considering all the eight genotypes, roots accumulated highest (51.18 %) portions of total As, and it was followed by shoots (stems + leaves + pods, 29%), pods (17.00 %) and distantly by seeds (2.82 %) (Table 1). Among the eight genotypes, L414 contained highest amount (89%) of total accumulated As in roots, and it was immediately followed by L 830 (86.67%). IPL 81, IPL 406 and DPL 59 contained 65%, 62.50% and 60% of total accumulated As in roots, respectively. Lowest accumulation (38.85%) was estimated in roots of local check cultivar (Table 1). In shoots (stems + leaves+ pods), highest accumulation (52.00%) was estimated in local check, and it was followed by VLM 4 (35.84 %), DPL 59 (35%), VLM 1 (32.83 %), IPL 81 (30%), IPL 406 (28.50%). Lowest As level (11%) in shoot was detected in L 414. In the edible part, highest As level (16.99%) was estimated in seeds of VLM 4, and was closely followed by VLM 1 (16.62%) and local check (9.15%). Seeds of IPL 81, IPL 406, DPL 59 and L 830 accumulated 5%, 9%, 5% and 0.33% of total accumulated As, respectively (Table 1). No As was detected in seeds of L 414. Among the treated samples, the BFs was the highest (0.31) in case of local check cultivar, and it was followed by VLM 4, IPL 406, VLM 1, DPL 59, IPL 81, L 830 and L 414 (Table 1). The BCFs was the highest (0.51) in L 414 and the lowest value (0.13) was recorded in IPL 406. Within this range, BCFs of 0.45 was recorded in L 830, and it was followed by local check, VLM 4, VLM 1, IPL 81, DPL 59 and others (Table 1). Barring local check, TFs were estimated below 1.0 in rest of the cases. Among these seven genotypes, TFs was the highest in VLM 4 (0.75), and it was immediately followed by VLM 1, DPL 59, IPL 81 and IPL 406 and was distantly by L 830, L 414 (Table 1). EFs were 0 in L 414, but it was > 100.0 in VLM 1 and local check cultivars (Table 1). EFs of 89.40 were recorded in IPL 406, but it declined around 50.00 in VLM 4 (40.47) and below 50.00 in both IPL 81 (32.80) and DPL 59 (30.63). It was low in L 830 (Table 1). In control plants, all the parameters were estimated low. The differences among genotypes became significant for all the four parameters described above (Table 1).

Table 1

Arsenic (As) concentrations (mg kg^{-1}) and bioaccumulation (bioaccumulation factors- BFs, bioconcentration factors-BCFs, transfer factors-TFs, and enrichment factors -EFs) in eight genotypes of lentil (Lens culinaris Medik.).

	Genotypes[A]								
Parameters[B]	IPL 81	IPL 406	L 414	DPL 59	L 830	VL M 1	VL M 4	Local check	control
Soil As	110.19 ± 0.25	-	-	-	-	-	-	-	0.96 ± 0.08
Roots As	21.37 ±0.11 ^{c}	31.05 ± 0.06 ^{b}	56.16 ±0.18 ^{a}	18.40 ± 0.05^{c}	49.09 ± 0.11 ^{a}	20.32± 0.12 ^{c}	21.70 ±0.11 ^{c}	25.56± 0.03^{c}	1.08 ± 0.06^{d}
Stem As	5.11± 0.07 ^{c}	7.56 ± 0.15 ^{b}	3.88 ± 0.18 ^{e}	3.11± 0.18 ^{e}	2.67 ± 0.20 ^{f}	3.50 ± 0.2 ^{a}	4.56± 0.06 ^{d}	9.88 ± 0.12^{a}	0.18 ±0.10^{g}
Leaf As	3.89 ± 0.01 ^{d}	5.67 ± 0.05 ^{c}	3.00 ± 0.02 ^{d}	2.05 ± 0.02 ^{e}	4.12 ± 0.03 ^{d}	7.17 ± 0.01 ^{b}	5.19 ± 0.01 ^{c}	20.67± 0.01^{a}	0.51 ± 0.03^{f}
Pod As	0.87 ± 0.08 ^{d}	0.92 ± 0.06 ^{d}	0.06 ± 0.12 ^{a}	5.57 ± 0.16 ^{a}	0.58 ± 0.03 ^{e}	2.63 ± 0.09 ^{c}	6.61 ± 0.02 ^{a}	3.66 ± 0.01^{b}	0.01 ± 0.02^{f}
Seed As	1.64 ± 0.03 ^{c}	4.47 ± 0.07 ^{b}	0.00 ± 0.00^{e}	1.53 ± 0.10 ^{c}	0.19 ± 0.09 ^{d}	6.88 ± 0.08 ^{a}	2.72 ± 0.03 ^{d}	6.02 ± 0.01^{a}	0.01 ± 0.01^{e}
BFs (shoot As/soil As)	0.09 ± 0.02 ^{b}	0.13 ± 0.10 ^{b}	0.06 ± 0.11 ^{c}	0.10 ± 0.05 ^{b}	0.07 ± 0.09 ^{c}	0.12 ± 0.10 ^{b}	0.15 ± 0.11^{b}	0.31 ± 0.08 ^{a}	0.006 ± 0.03^{d}
BCFs (Root As/ soil As)	0.19 ± 0.03 ^{b}	0.13 ± 0.05 ^{b}	0.51 ± 0.02 ^{a}	0.17 ± 0.05 ^{b}	0.45 ± 0.02 ^{a}	0.18 ± 0.02 ^{b}	0.20 ± 0.09 ^{b}	0.23 ± 0.09 ^{b}	0.01 ± 0.02 ^{c}
TFs (shoot As/root As)	0.46 ± 0.09 ^{c}	0.46 ± 0.11 ^{c}	0.12 ± 0.17 ^{d}	0.58 ± 0.08^{b}	0.15 ± 0.16 ^{d}	0.65± 0.02^{b}	0.75 ± 0.09^{b}	1.34 ± 0.11^{a}	0.65 ± 0.10 ^{b}
Seeds As (C)	0.05 ± 0.01	-	-	-	-	-	-	-	-
EFs	32.80± 0.10 ^{d}	89.40 ± 0.09 ^{b}	0.00 ± 0.00 ^{f}	30.63 ± 0.08 ^{d}	3.81 ± 0.09 ^{e}	137.6 ± 0.06 ^{a}	54.40 ±0.17 ^{c}	120.4 ±0.09^{a}	-

[A-] data are means ± SE of at least four replicates (n = 10), means followed by the same superscript letters are not significantly different at P < 0.05 by Duncan's Multiple Range Test. [B-] Seeds As (C)-As concentration in non-contaminated (control) locations (n = 10), EFs denotes the As concentration in the edible part (seeds) of plants grown in As-contaminated areas divided by As concentration in seeds of plants grown in non-contaminated (control) areas

DISCUSSION

The bioaccumulation potential of As in eight different genotypes of lentil was studied in pot experiment under controlled environmental conditions. Results in table 1 showed significant differences between the plant parts and also among eight genotypes regarding the accumulation and transport of As. Compared to control, the genotypes grown in As-treated soils exhibited significant accumulation of As in different plant parts, and the largest amount was measured in roots. This was evidenced by comparatively lower BFs values than BCFs in all the genotypes, except in local check where BFs exceeded BCFs value. Low BFs and high BCFs were mainly due to comparatively higher accumulation of As in the roots than that in the shoots. The result has immense significance as many plants avoid As-toxicity by limiting As-transport to shoot and increasing As accumulation in the root system [6], and lentil seedlings reportedly exhibited growth retardation under metal toxicity [19]. However, in IPL 406 both BFs and BCFs were similar, suggesting efficient translocation of As to its shoots in comparison to other genotypes. The results strongly indicated that lentil genotypes extracted significant amount of soil As through roots and transported it upward but in different magnitudes in eight genotypes. This differential translocation was reflected by variations in TFs among genotypes. High TFs values, especially in DPL 59, VLM 1, VLM 4 and local check suggested that these four genotypes transported a significant percentage of root As to shoots, varying between 0.58 (DPL 59) and 1.34 (local check). TFs value > 1.0 revealed over-accumulation of As in shoots compared to roots in local check cultivar due to extremely high transfer potential and suggested genotypic differences of locally adapted cultivar with improved genotypes for As accumulation and transport. In the present study, stems, leaves and pods were together considered as shoots. Results in table 1 indicated marked differences in As content among parts of shoots. The order (high to low) of As accumulation in shoot parts as stems>leaves>pods was found in three genotypes, namely IPL 81, IPL 406 and L 414 while the order of leaves>stems>pods were found in L 830, VLM 1 and local check. A completely different pattern pods> stems>leaves was observed in DPL 59 whereas pods>leaves>stems was manifested in VLM 4. High amount of As accumulation in the stems and leaves of above six genotypes indicated exposure of photosynthetic part to As in a significant way, but its distribution is somehow limited to reproductive organs. By contrast, high As level in pods of DPL 59 and VLM 4 in comparison to stems and leaves suggested distribution of As from photosynthetic parts to reproductive parts. Presumably, nutrient transport to pods which usually act as a powerful sink co-transferred and distributed substantial amount of As from other portions of shoot, particularly from leaves to pods. In legumes, the differential transport of As within plant parts has been attributed to chelation of As by phytochelatin compounds and subsequent sequestration of As [5, 20]. Similar possibilities have been explored in a glutathione-deficient and a glutathione-overproducing mutant of grass pea, a close ally of lentil, under cadmium stress [21, 22]. Obviously, the differential accumulation and transport pattern of As in the present lentil genotypes may be linked with thiol metabolism and phytochelatins on which further study is needed.

The transport of As from roots to the shoots was also found distributed in seeds, the ultimate sink and the edible part of a grain legume like lentil. The accumulation of As in seeds was measured and expressed as EFs. No As was detected in seeds of L 414, the genotype in which maximum accumulation took place in roots and a gradual decline was observed in aboveground parts. Similarly, low EFs (3.81) in L 830 strongly suggested successful prevention of As accumulation in seeds of the genotypes. On the other hand, the EFs > 100.0 in VLM 1 indicated substantial accumulation of As in treated plants compared with control, and the performance nearly corresponded local check cultivar. High to moderate enrichment of

seeds by As was also observed in rest of the genotypes, indicating presence of As in considerable amount in edible grain of the legume. Accumulation of As in seeds of pulse crops like lentil may have serious consequences in relation to nutritional quality of edible part [23], as seeds of this crop has been utilized in diverse types of food preparation, medicinal and pharmacological purposes [24, 25].

Accumulation of As is known to inhibit plant growth by inducing toxicity in root and shoots, as observed in different crops and legumes including chickpea, grass pea, and beans [3,4,5,26]. In grass pea, As treatment caused significant accumulation of As in the roots rather than in the shoots [3]. In lentil, importance of root and nodulation in plant growth and seed yield has been studied in presence of different abiotic stress factors including drought, salinity and heavy metals [11, 12]. On the other hand, As accumulation reportedly inhibited plant growth and seed yield in beans through impairment in photosynthetic apparatus [4, 26, 27]. In the present study, both roots and shoots accumulated As, with a possible effect on mineral nutrition and photosynthesis. Mitigation of As stress in lentil has been studied with mycorrhizal fungi [14], while application of calcium was found effective in preventing cadmium-induced oxidative stress in this crop [28]. The present results indicated sensitivity of lentil to metalloid toxicity which has also been manifested in diverse cellular and metabolic alterations in both leguminous and non-leguminous crops under stress [29,30,31,32,33].

CONCLUSION

The present results revealed that lentil genotypes grown in As-contaminated areas exhibited considerable variations in As accumulation and transport potential, although no genotypes can be considered as As hyper-accumulators due to values of BFs and BCFs <1.0. Comparing all the eight genotypes, L 414 and L 830 are relatively safe for edible purposes but it is noteworthy that As accumulation in photosynthetic part may jeopardize the food and nutritional quality of lentil grains, besides hampering its yield. Present investigation, for the first time, gives important first-hand information about the level of As in different parts of lentil crop which can be utilized for its future breeding programs. However, detail morpho-physiological and biochemical parameters are required in understanding of As-effect on cellular and metabolic responses of lentil crops towards better food and nutritional security of millions of people inhabiting As-prone zones and utilizing this pulse crop as the sole source of proteins in their daily diets.

REFERENCES

1. Sharma I, Travlos IS, Phosphate supply as a promoter of tolerance to arsenic in pearl millet. Int J Plant Prod, 6: 443-456, (2012).
2. Meharg AA, Macnair MR, Suppression of the high affinity phosphate uptake system: A mechanism of arsenate tolerance in *Holcus lanatus* L. J Exp Bot, 43: 519-524, (1992).
3. Talukdar D, Effect of arsenic-induced toxicity on morphological traits of *Trigonella foenum-graecum* L. and *Lathyrus sativus* L during germination and early seedling growth. Curr Res J Biol Sci, 3: 116-123, (2011a).
4. Talukdar D, Arsenic-induced oxidative stress in the common bean legume, *Phaseolus vulgaris* L. seedlings and its amelioration by exogenous nitric oxide. Physiol Mol Biol Plants, doi: 10.1007/s12298-012-0140-8, (2012d).
5. Gupta DK, Tripathi RD, Mishra S, Srivastava S, Dwivedi S, Rai UN, Yang XE, Huanj H and Inouhe M, Arsenic accumulation in root and shoot *vis-a-vis* its effects on growth and level of phytochelatins in seedlings of *Cicer arietinum* L. J Environ Biol, 29: 281-286, (2008).
6. Rahman F, Naidu R, The influence of arsenic speciation (As^{III} & As^{V}) and concentration on the growth, uptake and translocation of arsenic in vegetable crops

(silverbeet and amaranth): greenhouse study. Environ Geochem Health, 31: 115–124, (2009).

7. Cubero J I, Pérez de la Vega M, and Fratini R. Origin, phylogeny, domestication and spread. In: W. Erskine, F. J. Muehlbauer, A. Sarker, and B. Sharma (eds.), *The lentil: Botany, production and uses*, Comm. Agric. Bureau Int., Wallingford, 2009,pp. 13-33.
8. Erskine W, Sarker A and Kumar S. Crops that feed the world 3. Investing in lentil improvement toward a food secure world. Food Sec, 3:127-139, (2011).
9. FAOSTAT.Available at: http://faostat.fao.org, (2008).
10. Akibode S, Maredia M, Global and regional trends in production, trade and consumption of food legume crops, Draft Report, March 27, (2011).
11. Bandeoğlu E, Eyidoğan F, Yücel M and Öktem HA, Antioxidant responses of shoots and roots of lentil to NaCl-salinity stress. Plant Growth Regul, 42:69-77, (2004).
12. Muehlbauer FJ, Cho S, Sarker S, McPhee KE, Coyne CJ, Rajesh PN and Ford R, Application of biotechnology in breeding lentil for resistance to biotic and abiotic stress. Euphytica, 147:149-156, (2006).
13. Tepe M, Aydemir T, Antioxidant responses of lentil and barley plants to boron toxicity under different nitrogen sources. Afr J Biotechnol,10: 10882-10891, (2011).
14. Ahmed FR, Sadeque Killham K and Alexander I, Influences of arbuscular mycorrhizal fungus *Glomus mosseae* on growth and nutrition of lentil irrigated with arsenic contaminated water. Plant Soil, 258: 33-41, (2006).
15. Bhattacharya P, Samal AC, Majumdar J and Santra, SC, Arsenic contamination in rice, wheat, pulses, and vegetables: A study in an arsenic affected area of West Bengal, India. Water Air Soil Pollut, 213: 3-13, (2010).
16. Mascher R, Lippmann B, Holzinger S and Bergmann H, Arsenic toxicity: effects on oxidative stress response molecules and enzymes in red clover plants. Plant Sci, 163: 961-969, (2002).
17. Huang ZC, Chen TB, Lei M, Liu YR and Hu TD, Difference of toxicity and accumulation of methylated and inorganic arsenic in arsenic-hyperaccumulating and -hypertolerant plants. Environ Sci Technol, 42: 5106–5111, (2008).
18. Singh R, Singh DP, Kumar N, Bhargava SK and Barman SC, Accumulation and translocation of heavy metals in soil and plants from fly ash contaminated area. J Environ Biol, 31: 421-430, (2010).
19. Gautam S, Pandey SN, Growth and biochemical responses of nickel toxicity on leguminous crop (*Lens esculantum*) grown in alluvial soil. Res Environ Life Sci, 1: 25-28, (2008).
20. Flora SJS, Bhadauria S, Kannan GM and Singh N, Arsenic induced oxidative stress and the role of antioxidant supplementation during chelation: A review. J Environ Biol, 28: 333-347, (2007).
21. Talukdar D, A glutathione-overproducing mutant in grass pea (*Lathyrus sativus* l.): alterations in glutathione content, modiofications in antioxidant defense response to cadmium stress and genetic analysis using primary trisomics. Int J Recent Scientific Res, 3(4): 234-243, (2012c).
22. Talukdar D, An induced glutathione-deficient mutant in grass pea (*Lathyrus sativus* L.): Modifications in plant morphology, alterations in antioxidant activities and increased sensitivity to cadmium. Bioremediation, Biodiversity and Bioavailability, 6(spl issue 1): 75-86, (2012b).
23. Samal A C, Arsenic contamination in rice, wheat, pulses and vegetables: A study in an arsenic affected area of West Bengal, India. Water Air Soil Pollut, 213: 3-13, (2010).
24. Talukdar D, Talukdar T, Traditional legumes in Sikkim Himalayas: Food preparation, uses and ethno-medicinal perspectives. Int J Curr Res, 4(4): 64-73, (2012a).
25. Talukdar D, Talukdar T, Floral diversity and its indigenous use in old basin (Khari) of river Atreyee at Balurghat block of Dakshin Dinajpur district, West Bengal. NeBIO, 3(2): 26-32, (2012b).

26. Singh HP, Batish DR, Kohli RK and Arora K, Arsenic-induced root growth inhibition in mung bean (*Phaseolus aureus* Roxb.) is due to oxidative stress resulting from enhanced lipid peroxidation. Plant Growth Regul, 53(1): 65-73, (2007).
27. Stoeva N, Berova M and Zlatev Z, Effect of arsenic on some physiological parameters of bean plants. Biol Plant, 49(2): 293-296, (2005).
28. Talukdar D, Exogenous calcium alleviates the impact of cadmium induced oxidative stress in *Lens culinaris* Medic. seedlings through modulation of antioxidant enzyme activities. Journal of Crop Science and Biotechnology, in press, (2012e).
29. Talukdar D, The aneuploid switch: Extra-chromosomal effect on antioxidant defense through trisomic shift in *Lathyrus sativus* L. Indian J Fund Appl Life Sci, 1(4): 263-273, (2011b).
30. Talukdar D, Ascorbate deficient semi-dwarf *asfL1* mutant of *Lathyrus sativus* exhibits alterations in antioxidant defense. Biol Plant, 56: 675-682, (2012a).
31. Singh IK, Singh A, Rai P, Rocky I and Mangangcha, Role of mitochondrial antioxidants in tolerance against radiation stress in plants. Journal of Natural Science, Biology and Medicine, 2(2): 156, (2011).
32. Jayalakshmi NR, Thara Saraswathi KJ, Vijaya B, Raman DNS and Suresh R, Establishment of enhanced anthocyanin production in *Malva sylvestris* L. with different induced stress. International Journal of Pharma and Bio Sciences, 3(4): 17-27, (2012).
33. Jayachitra A, Padma PR, Antioxidant potential of Clitoria ternatea leaf extracts in vitro. International Journal of Pharma and Bio Sciences, 3(4):753-763, (2012).