Effects of Arsenic concentrations on growth and arsenic accumulation in *Eleocharis macrostachya, Shoenoplectus americanus and Baccharis salicifolia*

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Abstract
The effects of arsenic (As) on *Eleocharis macrostachya*, *Shoenoplectus americanus* and *Baccharis salicifolia* were investigated in a greenhouse study. The measured characteristics in the plant behavior were: As tolerance (growth and reproduction) and its accumulation. Plants were grown for a period of 10 weeks in arsenic-contaminated soil and water, at five different As concentrations (1.0. 2.0, 3.0, 4.0 and 5.0 mg/l). *E. macrostachya and S. americanus* behaved as tolerant plant with the highest biomass growth rate and arsenic uptake (up to 795 mg/kg). A symbiotic association of plant root with green algal species was observed in both plants. Arsenic translocation factors (TF) ranged from 0.4 to 8.96 in water (hydroponic) and from 0.1 to 4.8 in soil. *B. salicifolia* showed the highest (TF), but the greatest damage in plants. This research confirms the capability of *E. macrostachya and S. americanus* in arsenic phytoremediation.

Introduction
Arsenic (As) contamination has received widespread attention owing to its high toxicity to plants, animals and humans. In particular As contamination in groundwater has become a crucial environmental and health problem in some parts of the world as Bangladesh, Vietnam, India, China and Mexico (Milton et al., 2005). Huang et al. (2004) showed that the As hyperaccumulating ferns *Pteris vittata* and *Pteris cretica* were very effective in removing As from the water in a hydroponic system. A recent study has investigated the As accumulation potential of *Prosopis sp*. (Haque et al, 2009). The need for plant species to sustain growth at contaminated sites is one of many phytoremediation limitations. Thus, selection of plant species for phytoremediation is an ongoing process. Plant biomass production and plant elemental uptake are two key factors for successful application of phytoremediation (Reeves and Baker, 2000). *E. macrostachya, S. americanus y B. salicifolia* are three species of native plants in the state of Chihuahua Mexico, that have been reported as arsenic tolerant (Alarcón et al 2009). The objectives of this research were to determine the impacts of As toxicity on plant biomass and growth of *E. macrostachya, S. americanus* and *B. salicifolia*; and determine the capacity of these plants on arsenic uptake.

Materials and Methods
Experiment setup
The experiment was performed considering the plants exposure to 5 different arsenic concentrations (1.0, 2.0, 3.0, 4.0 and 5.0mg/l) and tap water as control (0.016mg/l). The experiment was carried out with soil (sandy loam) as plants support and without soil (hydroponic conditions, without nutrients). At the end of the experiment, the total arsenic concentration in plants and soil was quantified.

Effects of arsenic concentrations on plants growth
To determine the effect of arsenic concentration on plant growth, three plants were randomly selected and the size of the roots and shoots were measured in each pot, for each level of As concentration (90 plants). Each plant was measured from the main apex of the root to the crown and from the crown to the main apex of the shoot. All plants were carefully prepared and weighed at the beginning and the end of the experiment to determine plants biomass.

Arsenic quantifications in plants and soil
The plants were harvested after 10 weeks of growth, separated into stem and roots, oven-dried at 60°C, and then ground into powder. Soil and plant samples were digested in a microwave oven and analyzed for total As, using an inductively coupled plasma optical emission spectrophotometer (ICP-OES).

Results
At the end of the experiment, *S. americanus* presented the highest arsenic accumulation with a maximum concentration of 795.11mg/kg **(Figure 1)**.

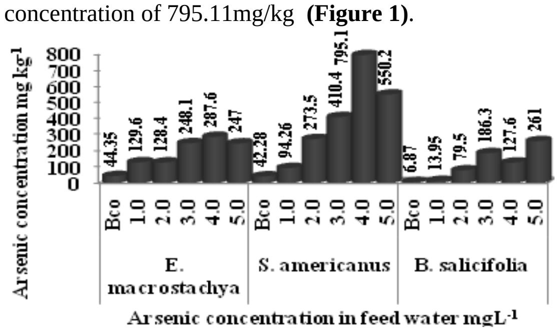

Figure 1.-Total arsenic concentration (mg/kg) in *E. macrostachya*, *S. americanus* and *B. salicifolia* after growing for 10 weeks in hydroponic conditions.

E. macrostachya and *B. salicifolia* showed cellular tissue damage (chlorosis and necrosis) in their aboveground biomass after three weeks of being subjected to arsenic concentrations above 2.0mg/l. During the time of the experiment, in all arsenic concentrations, *S.* americanus showed no evidence of physical damage to cell tissue or limitation on reproduction or growth. Translocation factor, plant biomass and plant growth are presented in table 2.

Table 2 .- Effect of As concentration on plant biomass and growth of *E. macrsotachya, S. americanus* and *B. salicifolia* using soil as medium of support.

	As treatment (mgAs/l)	Biomass(g)	Growth (cm)	TF
E. macrostachya	Control(0.016)	65.96	112	0.57
	1.0	58.81	64	0.39
	2.0	32.97	100	1.64
	3.0	95.71	49	1.86
	4.0	65.98	93	0.79
	5.0	84.19	75	0.99
S. americanus	Control(0.016)	86.26	76	0.32
	1.0	158.63	75	1.34
	2.0	110.20	45	0.07
	3.0	155.76	85	0.10
	4.0	149.12	90	0.47
	5.0	191.28	61	1.73
B. salicifolia	Control(0.016)	17.63	31	0.22
	1.0	19.65	31	3.31
	2.0	26.97	41	4.79
	3.0	18.06	3	0.21
	4.0	15.21	32	0.54
	5.0	33.10	50	2.05

Discussions

During the experiment, *S. americanus* and *E. macrostachya* decreased their ability to absorb arsenic at arsenic concentrations upper to 4.0 mg/l (Figure 1). This fact could be attributed to a plants defense mechanism to avoid damage of cellular tissue. *E. macrostachya* behaved as traslocator at arsenic concentrations above 2mg/l, which agrees with the detection of the damage in the cellular tissue, observed in the aboveground biomass. Both plants, developed longer roots at higher arsenic concentrations, while *B. salicifolia* had an opposite behavior.

B. salicifolia had the highest TF in both soil and water (4.79 and 8.96 respectively), but the greatest damage in plants. This plant showed the lowest growth and arsenic uptake. Other plants like Arundo donax, a tall perennial cane growing in damp soils, either fresh or moderately saline presented a TF of 4.93 when was subject to arsenic concentration of 1.0mg/l (Mirza et al., 2010). The plant is an As traslocator as *B. salicifolia,* but A. donax doesn't present any damage.

E. macrostachya and S. americanus showed TF <1 in most of the concentrations of As tested, which is typical of non-hyperaccumulator plants.

Another interesting feature of the present study, was the symbiotic association between *E. macrostachya* and *S. americanus* roots with some green algal species. In spite of high arsenic concentrations, the growth of these so-called symbiotic algae could not be inhibited. Thus, the algal biomass was also analyzed to know the total arsenic concentration; it was shown that arsenic was also accumulated in the algal biomass (381mg/kg). *B. salicifolia* did not show this association.

Conclusions

The analyzed plants have a higher susceptibility to As toxicity under hydroponic conditions.

E. macrsotachya and *S. americanus* have a high arsenic tolerance. The symbiotic association of plant root with green algal species could be an explanation for the greatest arsenic tolerance. Both species could be use in rhizofiltration and phytostabilization techniques for the remediation of contaminated water.

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