Phytoaccumulation Potential and Toxicity of Arsenic Ions by *Eichhornia Crassipes* in Hydroponic System

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**Abstract**

In this work, the phytoaccumulation efficiency of total arsenic (As) by the aquatic plant *E. crassipes* has been studied. Plants were cultured in double distilled water with modified 0.25 N Hoagland’s nutrient solution at pH 6.8 supplemented with 0, 0.010, 0.025, 0.05, and 0.10 mg L⁻¹ equally mixing (1:1) arsenic (III) and arsenic (V). They were harvested after 0, 3, 9 and 15 days. Plants treated with 0.10 mg L⁻¹ arsenic solutions, accumulated the highest concentration in roots (7.2 mg kg⁻¹, dry weight) and shoots (32.1 mg kg⁻¹, dry weight) respectively after 15 days. SEM-EDX and FTIR were used to characterize the interaction between arsenic ions and extract materials of *E. crassipes* shoot biomass. The FTIR spectra indicated the binding characteristics of the arsenic ions involved the hydroxyl, amide, amino and thiol groups in the biomass. The relative growth and bio-concentration factor of plants at different concentration of arsenic solutions significantly increases (P < 0.05) with the passage of time. From the above studies it is clear that the plant *E. crassipes* can be used successfully for the removal of arsenic ions by phytoremediation process.

**Keywords:** Phytoremediation; *Eichhornia crassipes*; Total arsenic; SEM-EDX; FTIR

**Introduction**

Rapid industrialization and urbanization have caused elevated release of toxic heavy metals which ultimately increase the load of pollutant in the biosphere [1]. Recently considerable interest is growing to develop cost effective and environmental friendly technologies for the remediation of toxic metals from waste water with the help of cultured plants; this technique is known as phytoremediation [2,3]. *Eichhornia crassipes* is common macrophyte which is abundant in wetlands, lakes and ditches. As it has a high growth-rate, fibrous root system and broad leaves along with tendency to tolerate high metal concentration, it is considered as an important species to be used in phytoremediation [4,5]. Some macrophytes are found to remove different concentrations of arsenic ions, which make them suitable to act as bio-monitors for metals, and have ability to act as biological filters of the aquatic environment [6-9]. Arsenic is one of the most significant and priority pollutant which is found in natural and anthropogenic processes like weathering, biological activity, geochemical reactions, volcanic eruption, smelting of non-ferrous metals and burning of fossil fuels [10]. Toxicity of arsenic to plants depends on its valence state; As (III) is 60 times more toxic, soluble and mobile than As (V), less toxic than As (VII), which usually occurs associated with oxygen as arsenate (AsO₄³⁻, HAsO₄²⁻, H₂AsO₄⁻) oxyanions [11,12]. Therefore, World Health Organization (WHO) has recommended the permissible concentration of arsenic in drinking water should be 10 µg L⁻¹ [13]. Various remediation methods exist that include chemical, physical, and biological technologies. Among these methods, use of aquatic plants to absorb metals from surrounding water is extremely efficient.

A plant-based phytoremediation approach to heavy metals uses plant roots to extract, the vascular system to transport and the leaves as a sink to concentrate the elements above ground for harvest and processing. The use of hydroponic culture treatment has been suggested as a means of assessing the plant tolerance to the toxic elements or its efficiency in mineral utilization. The bio removal process using aquatic plants contains two uptake processes such as (i) biosorption which is an initial fast, reversible, and metal-binding process and (ii) bioaccumulation which is a slow, irreversible, and ion-sequestration step. The major advantages of this technique are the environmentally friendly, cost-effective, aesthetically pleasing, technologically feasible, long-term applicability, and ecological aspect [14]. Many results have been documented the phytoremediation ability of the free-floating *E. crassipes* for nutrient-rich waters [15,16].

The aim of the present study was (i) to evaluate the uptake of arsenic ions (mg kg⁻¹) with different concentration by *E. crassipes* from aqueous solution. (ii) To study the phyto-accumulation potential of *E. crassipes*, relating growth, bio-concentration factor and toxicity of treated plants are also studied. (iii) Further *E. crassipes* biomass is characterized by SEM-EDX and FTIR techniques to know the absorption of arsenic ions.

**Materials and Methods**

**Experimental procedure**

Young aquatic plants *Eichhornia crassipes* were collected from ponds near civil township, Rourkela. The plants were placed in cement tanks with tap water under natural sunlight for one week to allow them to adapt to the new environment, and then the each plant was approximately weighed 250g of 10 to 11 cm root length and 6 to 7 cm shoot length were selected for further experimentation. The modified 0.25 N Hoagland nutrient solution are prepared which consists of 4.0 mM Ca(NO₃)₂, 2.0 mM MgSO₄, 4.0 mM KNO₃, 0.4 mM (NH₄)₂SO₄, 2 µM MnSO₄, 0.3 µM CuSO₄, 0.8 µM ZnSO₄, 30 µM NaCl, 0.1 µM Na₂Mo₄, 1.43 µM KH₂PO₄, 10 µM H₂BO₃, and 20 µM Fe-Na-EDTA (All obtained SEM-EDX; FTIR; 0.010, 0.025, 0.05, and 0.10 mg L⁻¹ equally mixing (1:1) arsenic (III) and arsenic (V). They were harvested after 0, 3, 9 and 15 days. Plants treated with 0.10 mg L⁻¹ arsenic solutions, accumulated the highest concentration in roots (7.2 mg kg⁻¹, dry weight) and shoots (32.1 mg kg⁻¹, dry weight) respectively after 15 days. SEM-EDX and FTIR were used to characterize the interaction between arsenic ions and extract materials of *E. crassipes* shoot biomass. The FTIR spectra indicated the binding characteristics of the arsenic ions involved the hydroxyl, amide, amino and thiol groups in the biomass. The relative growth and bio-concentration factor of plants at different concentration of arsenic solutions significantly increases (P < 0.05) with the passage of time. From the above studies it is clear that the plant *E. crassipes* can be used successfully for the removal of arsenic ions by phytoremediation process.

**Keywords:** Phytoremediation; *Eichhornia crassipes*; Total arsenic; SEM-EDX; FTIR
from Merck Specialties) [17]. The experiments were conducted in a series of rectangular glass container (32 x 20 x 18 cm) containing 200 mL/L of nutrient solutions. Each rectangular glass container was kept with two liters of deionized water and 3 numbers of selected plants in a growth room at 25 ± 2°C lighted with cool, fluorescent lamp (200 μmol.m⁻².s⁻¹) under 16 h photoperiod. The pH (Orion two stars, USA) of the nutrient solution was adjusted to 6.8-7.2 using required amount of 0.1 M HCl or 0.1M NaOH. This range of pH of the nutrient solution is most suitable for best growth of aquatic plants. The solution was changed regularly at intervals of 3 days to maintain the desired pH. The plants were maintained in the rectangular glass container supplemented with 0, 0.010, 0.025, 0.05, and 0.10 mg.L⁻¹ equally mixing (1:1) arsenic (III) and arsenic (V) solutions to expose the plants. They were harvested after 0, 3, 9 and 15 days treatments each container collected one plant. Deionised water was added daily to compensate the water loss through plant transpiration, sampling and evaporation. After the completion of each test duration, E. crassipes were separated into shoots and roots for the analysis of relative growth, metals accumulation, toxicity and bio-concentration factor. In addition, the residual arsenic ions in the solution were measured to assess the removal potential of E. crassipes.

Preparation of standards and reagents
All the chemicals used in the studies are of analytical grade which are used without further purification. In all experiments, deionized water (Milli-Q Millipore 18.2 MΩ cm⁻¹ conductivity) is used for the preparation, dilution and analytical purposes of solutions. 1000 mg.L⁻¹ of arsenic (III) stock solution is prepared by dissolving 1.320 g of arsenite (As₂O₃; Merck Chemicals, Germany) in deionized water containing 4 g 1M NaOH in 1 L of double distilled water. 1000 mg.L⁻¹ of arsenate (V) stock solution prepared by dissolving 4.164 g of sodium arsenate (Na₃H₂AsO₄·7H₂O; Merck Chemicals, Germany) in 1 L of double distilled water [18]. The stock solutions are preserved with 1% trace metal grade nitric acid. Subsequently, different working arsenic solutions of required concentrations are prepared by proper dilution. The 500 mL NaBH₄ solution is prepared by dissolving 2.5 g NaOH and 2.0 g NaBH₄ in double-distilled water and diluting up to mark. The NaBH₄ reagent is always prepared immediately before use. Sodium tetrahydrosborate solution is dispensed into the acidic test sample solution. The reaction of sodium tetrahydrosborate in acidic solution and the simultaneous reduction of the hydride-forming element can be described as represented.

\[
\text{BH}_4^- + \text{H}_2\text{O} \rightarrow \text{H}_3\text{BO}_3^- + 4\text{H}_2\text{O} + 4\text{H}_3\text{O}^+ \\
3\text{BH}_4^- + 3\text{H}^+ + 4\text{AsO}_3^2^- \rightarrow 4\text{AsH}_3^+ + 3\text{H}_2\text{O} + 3\text{H}_3\text{BO}_3
\]

Analytical methods
Water samples of 10 mL were taken daily from the container in order to measure the removal of arsenic ions from the nutrient solution. The freeze-dried samples were ground to fine powder using a ceramic mortar and pestle, and about 0.5 g was weighed into a 50 mL conical flask to which 10 mL of concentrated nitric acid was added. The mixture was then heated at 50°C for 30 min, than the temperature was further increased to 70°C. When the volume of acid decreased to 1 mL, the flasks were allowed to cool to room temperature and made up to a final volume (10 mL) with Milli-Q water. In each analytical batch, at least 2 reagent blanks and one internationally certified reference material (2 replicates) were included. To measure total arsenic concentration in the above digested samples, 1 mL of digest was mixed with 9 mL of reducing solution consisting of 1.5% (w/v) potassium iodide, 1.5% (w/v) ascorbic acid and 10% (v/v) hydrochloric acid. This mixture was then heated at 50°C for 1h. Total arsenic in digests was determined by a HG-AAS (Perkin-Elmer P 200, USA). The carrier solution was 5% (v/v) nitric acid, and the reductant solution consisted of 0.2% (w/v) sodium borohydride and 0.05% (w/v) sodium hydroxide. The results of the accumulation were reported as concentration (mg kg⁻¹), dry weight of arsenic ions in plants.

The relative growth of plant species is a major factor for contributing to invasion. Relative growth of treated plants was calculated as follows [19,20] and the results are expressed as increases of biomass per unit mass per day (g.g⁻¹.d⁻¹).

Relative growth = Final fresh weight / Initial fresh weight

The BCF provides an index of the ability of the plant to accumulate the metal with respect to the metal concentration in the substrate. The result of BCF was calculated as follows [21].

BCF= Concentration of metal in plant tissue (mg kg⁻¹) / Initial concentration of metal in external solution (mg L⁻¹).

The mean of relative growth, bio-concentration factor, accumulation and arsenic ions extraction efficiency were calculated and subjected to analysis of variance test by using MINTTAB 15 software [22].

Results and Discussion
Relative growth
The effects of arsenic ions on relative growth of E. crassipes at different concentrations and exposure times were presented in Table 1. The relative growth of control plants significantly increased (P < 0.05) with the passage of time. In plants treated with arsenic, the relative growth significantly increased (P < 0.05) in 0.010, 0.025, 0.05 and 0.10 mg.L⁻¹ treatments. All the statistical tests have been conducted at a level of 0.05 levels and presented in Table 2. The highest values of relative growth were 1.34 g.g⁻¹.d⁻¹ for E. crassipes treated with arsenic ions at 0.1 mg.L⁻¹. The best way of long term strategy for improving phytoextraction is to understand and exploit the biological processes involved in metal acquisition, transport and shoot accumulation. This is due to higher concentrations level of heavy metal ions have inhibitory effects on plant metabolic activity, alternatively reduced growth of plants, leaf necrosis and inhibits the plant physiology systems [8].

Bio-concentration factor
The bio-concentration factor values of arsenic ions at different

<table>
<thead>
<tr>
<th>Arsenic ions con. mg/L</th>
<th>Mean ± S.D.</th>
<th>Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.010</td>
<td>1.06 ± 0.01</td>
<td>1.14 ± 0.05</td>
</tr>
<tr>
<td>0.025</td>
<td>1.11 ± 0.03</td>
<td>1.25 ± 0.03</td>
</tr>
<tr>
<td>0.050</td>
<td>1.23 ± 0.02</td>
<td>1.26 ± 0.04</td>
</tr>
<tr>
<td>0.10</td>
<td>1.25 ± 0.01</td>
<td>1.30 ± 0.02</td>
</tr>
</tbody>
</table>

S.D. = Standard deviation
The standard deviation has been obtained for n=3

Table 1: The effects relative growth and bio-concentration factor of E. crassipes at different arsenic ions concentration and exposure times.
concentration and exposure times were also presented in Table 1. Bioconcentration factor is a useful parameter to evaluate the potential of the plants in accumulating metals. The bio-concentration factor for arsenic ions increases with the passage of time. As expected the bio-concentration factor for arsenic ions significantly increased (P < 0.05). The arsenic concentration in the feed solution at each exposure time for this test are presented in Table 2. The maximum bio-concentration factor values for arsenic were 323, indicating that E. crassipes can be used for effective phytoremediation. It is reported that the bio-concentration factor values of E. crassipes are very high for Cd, Cu, Cr and Se at low external concentration, and they are found to decrease with the increase in external concentration [21,23]. It is inferred that this is a cost-effective plant-based technology for the removal of toxic metals from the environment and has great potential for future applications.

### Arsenic ions translocation and accumulation mechanism

Arsenic ions accumulation by E. crassipes at different concentrations and exposure times were presented in Figure 1. From the data in Table 3 it is inferred that, there was an increase in the arsenic ions accumulation in shoots and roots when arsenic concentration and exposure times were increased (P < 0.05) which are presented in Table 4. Table 1 shows the accumulation capacity of arsenic ions in different parts of plants. Plants treated with 0.10 mg L⁻¹ of arsenic for 15 days accumulated the highest arsenic ions in shoots (32.1 mg kg⁻¹ dry weight) and in roots (9.2 mg kg⁻¹, dry weight). Similar results have been obtained in field studies for the accumulation of metals in the roots and shoots of E. crassipes [24,25]. Arsenic once accumulated inside the plant, the arsenic ions must be translocated through the xylem at high concentrations in a manner that does not disrupt cytoplasmic function, which gives the considerable phytotoxicity of arsenic species. Finally, the arsenic is stored at very high concentrations in the shoots. The metals accumulation in water hyacinth increased linearly with the solution concentration in the order of leaves > stems > roots in E. crassipes [15,21,26]. Metal ions penetrated plants by passive process, mostly by exchange of cations which occurred in the cell wall. All heavy metals were taken up by plants through absorption, translocation and released by excretion. It can be proposed that the roots reached saturation during the period and there exists some mechanism in roots that could detoxify heavy metals or transfer them to aerial parts [27].

The most efficacious remediation of arsenic requires that plants extract arsenic from water and accumulate in shoot parts. To accomplish this, the electrochemical species of arsenic must be changed in different parts of the plant. The bacterial arsenate reductase (ArsC) catalyzes the electrochemical reduction of arsenate to arsenite. The bacterial γ-glutamylcysteine synthetase (γ-ECS) catalyzes the formation of γ-glutamylcysteine (γ-EC) from the amino acids glutamate and cysteine and is the committed step in the synthesis of glutathione (GSH) and phytochelatines (PCs) (indicated by three arrows) [28]. Reduced arsenite can bind organic thiols (RS) such as those in γ-EC, GSH, and PCs through the replacement of oxygen by organic sulfur species. The scheme of reaction mechanism is as follows [29].

![scheme of reaction mechanism](image)

The fraction of the metal ions which has not been absorbed by the plants might be reacted with nutrient solution in container (Table 5). The mass balance of the arsenic ions is calculated in this process:

\[
\text{[Initial arsenic concentration in the container]} = \text{[Residual arsenic ions in the container]} + \text{[Accumulated arsenic ions in plants]}
\]

#### Arsenic ions detoxification

The toxic effects of arsenic ions mainly depend on the metal speciation, which decides its uptake, translocation and accumulation mechanism. A strategy for cells to detoxify non-essential metal ions and an excess of essential metal ions is the synthesis of high-affinity binding sites to suppress binding to physiologically important functional groups. Given the high reactivity of metal ions with thiol, amino or hydroxyl groups it is not surprising that molecules carrying these functional groups have been described as metal chelators. The best-known and presumably most effective chelators for arsenic ions are small Cys-rich proteins (metallothioneins, MTs) and Cys-containing peptides glutathione (GSH) and phytochelatines (PCs). PCs peptides of general structure (γ-Glu-Cys), Gly (n = 2-11) (Figure 2a) [28]. They are enzymatically synthesized by a specific transpeptidase, the phytochelatin synthase, which is activated by the presence of metal ions and uses glutathione as substrate. Their detoxifying function consists in the ability to bind metals in stable complexes, which effectively reduce the intracellular concentration of potentially toxic free metal ions. A detoxification pathway for arsenate (AsO₄³⁻) by conversion to arsenite (AsO₃²⁻) upon its uptake into roots has been proposed [28]. Arsenite (AsO₃²⁻) transformation and detoxification system in root cells is responsible for the phytochelatins and oxidized to form arsenate.

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**Table 2: ANOVA table for relative growth and bio-concentration factor.**

<table>
<thead>
<tr>
<th>Source</th>
<th>Degree of Freedom (df)</th>
<th>Sum of Squares (SS)</th>
<th>Mean Squares MS = SS/df</th>
<th>F-Statistics</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Relative growth</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Different concentration</td>
<td>3</td>
<td>0.000645</td>
<td>0.00215</td>
<td>20.59</td>
<td>0.001</td>
</tr>
<tr>
<td>Different days</td>
<td>2</td>
<td>0.003345</td>
<td>0.01053</td>
<td>16.53</td>
<td>0.004</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>0.00063</td>
<td>0.000104</td>
<td>13.2</td>
<td>0.012</td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>0.01053</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bio-concentration factor</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Different concentration</td>
<td>3</td>
<td>0.03612</td>
<td>0.012042</td>
<td>25.70</td>
<td>0.001</td>
</tr>
<tr>
<td>Different days</td>
<td>2</td>
<td>0.033008</td>
<td>0.016504</td>
<td>35.23</td>
<td>0.000</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>0.002811</td>
<td>0.00046</td>
<td>20.59</td>
<td>0.001</td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>0.071944</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

F = MSB/MSW. The between-sample variance, SS = SSB/n-1, where n is the number of observations in all the samples is often called the Mean Square Within, MSW = SSW/n-k, where n is the total number of observations in all the samples is often called the Mean Square Within or Mean Square Error, MSE = SSW/n-k.
Figure 1: The accumulation of arsenic ions at pH 6.8 in (a) shoots and (b) roots of *E. crassipes* biomass.

### Table 3: The accumulation of total arsenic (As) at pH 6.8 in shoots and roots stems *E. crassipes* at different arsenic ions concentrations and exposure times.

<table>
<thead>
<tr>
<th>Source</th>
<th>Degree of Freedom (df)</th>
<th>Sum of Squares (SS)</th>
<th>Mean Square (MS)</th>
<th>F-Statistics</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shoots</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Different concentration</td>
<td>3</td>
<td>2.30222</td>
<td>0.01904</td>
<td>532.5</td>
<td>0.000</td>
</tr>
<tr>
<td>Different days</td>
<td>2</td>
<td>0.03808</td>
<td>0.007641</td>
<td>12.98</td>
<td>0.007</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>0.00880</td>
<td>0.00147</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>2.34910</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Different concentration</td>
<td>3</td>
<td>1.2672</td>
<td>0.4224</td>
<td>3210.5</td>
<td>0.000</td>
</tr>
<tr>
<td>Different days</td>
<td>2</td>
<td>0.01778</td>
<td>0.00891</td>
<td>67.72</td>
<td>0.000</td>
</tr>
<tr>
<td>Error</td>
<td>6</td>
<td>0.00078</td>
<td>0.00013</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>1.28587</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Table 4: ANOVA table for arsenic ions absorption efficiency of shoots and roots.

<table>
<thead>
<tr>
<th>Arsenic solution (mg/L)</th>
<th>Arsenic accumulation (%) in roots</th>
<th>Arsenic accumulation (%) in shoots</th>
<th>Arsenic remaining (%) in container</th>
<th>Absorption efficiency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.010</td>
<td>14.11</td>
<td>47.70</td>
<td>36.19</td>
<td>61.81</td>
</tr>
<tr>
<td>0.025</td>
<td>14.72</td>
<td>51.96</td>
<td>33.32</td>
<td>66.68</td>
</tr>
<tr>
<td>0.050</td>
<td>13.04</td>
<td>62.00</td>
<td>24.96</td>
<td>75.04</td>
</tr>
<tr>
<td>0.100</td>
<td>17.03</td>
<td>68.93</td>
<td>14.04</td>
<td>85.96</td>
</tr>
</tbody>
</table>

Table 5: Mass balance of arsenic ions in various parts of *E. crassipes* after 15 days treatments.
Arsonate can be reduced to arsenite enzymatically by arsenate reductase as shown in vitro and non-enzymatically by glutathione (GSH) [30] followed by the formation of an arsenite-thiol (AsO$_3^{--}$-SH) complex. Acylation of binding site (Figure 2b) I (step 1) occurs at a cysteine that is 100% conserved in all known phytochelatin synthases and phytochelatin synthase-like proteins. The cysteine together with a histidine and an aspartate forms the catalytic triad typical for cysteine proteases. Glycine is cleaved off (step 2) and the resulting γ-glutamylcysteine dipeptide is transferred onto another glutathione (or a PC molecule). Binding site II remains to be identified and is possibly not present in bacterial phytochelatin synthase-like proteins [31]. They catalyse steps 1 and 2, resulting in the degradation of glutathione to γ-glutamylcysteine and glycine. Steps 1 and 2 are metal ion-independent. Acylation of site II and peptide transferase activity require metal ion activation and/or the binding of a metal-glutathione complex.

Characterizations of *E. crassipes* shoot biomass after absorption of arsenic ions

**SEM-EDX analysis:** The surface morphology of *E. crassipes* shoot biomass without and with removal of arsenic ions during absorption process is observed with the help of SEM-EDX (JOEL model JSM-6480LV, Japan) and is presented in Figure 3. (Figure 3a) clearly reveals the surface texture and pores in the materials. (Figure 3b) shows the morphological changes with respect to shape and size of the materials after absorption of arsenic ions. It can be clearly observed that the surface of extract materials shape has changed into a new shiny bulky particles and whitish patches structure after arsenic ions absorption. The EDX spectra of arsenic ions unloaded and loaded of extract materials obtained is shown in Figure 3a and Figure 3b, respectively. So, it is concluded that, arsenic ions are adsorbed on the surface of the extract materials. These results are further confirmed with the results of FTIR spectra analysis.

**FTIR analysis:** Infrared spectra (Perkin Elmer Spectrum RX-I) of the *E. crassipes* shoot biomass with and without arsenic loaded are obtained to determine which functional groups may have contributed to the arsenic absorption are presented in Figure 4(a) and Figure 4(b). The FTIR spectra of the shoot biomass without arsenic displays a number of absorption peaks, indicating the complex nature of the biomass. The spectra of loaded with arsenic and without are compared and found the following shift. The spectra of extract materials exhibits a broad absorption band at 3225.78 cm$^{-1}$ due to bonded – OH stretching vibration which is shifted to 3195.91 cm$^{-1}$ may be due to complexation of –OH groups. The band at 2,918.50 cm$^{-1}$ has been shifted insignificantly. The new peak at 2,351.75 cm$^{-1}$ may be due to the complexation of –SH group with arsenic ions [32].

The next absorption peak at 1,645.03 cm$^{-1}$ may be due to the presence of amide group (N-H stretching and C=O stretching vibration) is shifted to higher frequency to complexation of –OH groups. The band at 2,918.50 cm$^{-1}$ has been shifted insignificantly. The new peak at 2,351.75 cm$^{-1}$ may be due to the complexation of –SH group with arsenic ions [32]. The next absorption peak at 1,645.03 cm$^{-1}$ may be due to the presence of amide group (N-H stretching and C=O stretching vibration) is shifted to higher frequency and appeared at 1,645.03 cm$^{-1}$ may be due to the complexation of amide
group with arsenic ions [33]. Another peak at 1, 319.75 cm⁻¹ has been shifted insignificantly. Another shift was observed from 1,163.34 cm⁻¹ to 1,169.27 cm⁻¹ and 1,022.47 cm⁻¹ to 1,023.65 cm⁻¹ may be due the interaction of nitrogen from amino group with arsenic ions [34,35].

The other weak absorption peak shifted from 780.91 cm⁻¹ to 780.64 cm⁻¹, 670.09 cm⁻¹ to 668.44 cm⁻¹, 632.40 cm⁻¹ to 630.18 cm⁻¹ and 522.34 cm⁻¹ to 522.75 cm⁻¹ corresponding to the thiol or sulfhydryl group with arsenic ions [32]. The above changes in the spectra may be attributed to the interaction of arsenic ions with the hydroxy, amide, thiol and amino groups present on the shoot biomass.

Conclusions

From the above discussion it is concluded that hydroponic culture is an efficient method for screening of arsenic ions tolerant for free floating plants of E. crassipes. The maximum absorption efficiency of arsenic ions is 85.96 % after 15 days treatments. FTIR analysis reveals that the arsenic ions may be co-ordinated with the hydroxy, amide, thiol and amino groups present in the biomass. SEM- EDX analysis also confirms the absorption of arsenic ions in the plants. ANOVA analysis of relative growth, bio-concentration factor, and arsenic accumulation statically reveals P < 0.05. The high removal efficiency and more accumulation capacity of arsenic ions make E. crassipes an excellent choice for phytoremediation processes.

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References