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## RESPONSE OF ARUM (*COLOCASIA ANTIQUORUM*) TO DIFFERENT LEVELS OF ARSENIC (As) TREATMENTS

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### Abstract

An experiment was conducted with one variety Bilasi, and two germ-plasms - Mukhi-029 and Mukhi-140 of arum to compare their responses to different levels of arsenic. All these materials showed severe symptoms of As toxicity - reduced vegetative growth - at As application of 80 mg/l. Arsenic treatments had significant ( $p = 0.002$ ) negative effect on dry matter production of the individual material as well as As accumulation ( $p < 0.001$ ). Mukhi-029 accumulated the maximum amount of arsenic (334.33 mg/kg) followed by Mukhi-140 (209.73 mg/kg) and Bilasi (195.88 mg/kg). The As accumulation followed the order: Mukhi-029 > Mukhi-140 > Bilasi while the individual plant parts followed the order: root > stem > leaf irrespective of the variety or germplasm.

### Introduction

Groundwater arsenic and its fate in the environment has become a matter of great concern all over the world including Bangladesh.<sup>(1)</sup> More than 40 per cent of net cultivable land is under irrigation and more than 60 per cent of this irrigation is met from ground water.<sup>(2)</sup> Shallow ground water used for cultivation is also contaminated with arsenic and the source of contamination is geogenic.<sup>(3)</sup> Arsenic enters the living biota through biogeochemical and biochemical pathways.<sup>(4)</sup> Arsenic enters the human body through drinking water and contaminated food sources and becomes the causes for many chronic and acute health consequences. The principal sources of nourishment in the rural population are green vegetables and root crops. In rural areas, vegetables are grown in the homestead gardening where As contaminated water is used to irrigate them. The concentration of As has been observed to be particularly high in arum (*Colocasia antiquorum*) - locally known as "kochu", gourd leaf and in kalmi sak.<sup>(5)</sup> Moreover, Huq and Naidu<sup>(4)</sup> have observed that arum is an As accumulator. Incidentally, these are some of the most commonly used vegetables among the rural population. Every part of this plant: leaves, stems, rhizomes are edible and consumed. In Bangladesh, farmers cultivate seven varieties of arum, that include 'man kachu', 'pani kachu', 'gut kachu', 'kalika kachu', 'bish kachu', 'ole kachu' and 'panchamukhi kachu (locally known as mukhi),<sup>(6)</sup> and all of these are consumed by the people. 'Mukhi kachu' and 'Pani kachu' are most important and are commer-

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cially grown in all parts of the country. 'Mukhi kachu' (*Colocasia antiquorum*) of the family Araceae is one of the important edible aroids in Bangladesh. Among the tuber crops the corms and cormels of Mukhi kachu are rich source of carbohydrate, vitamins A and C, good source of iron and also contain sufficient quantity of protein.<sup>(7)</sup> It is extensively grown in Bangladesh in Kharif season with supplemental irrigation and contributes a considerable part in the total supply of bulky vegetables when other sources are scarce in market.<sup>(6)</sup> It is however, not known about the difference among arums in respect of As accumulation and dynamics. It would be prudent to avoid the accumulator arum from cultivation with As contaminated water. With this view in mind, the present experiment was conducted with one variety and two germplasm of arum to see their relative As accumulation capacity.

### Materials and Methods

The experiment was conducted in the Department of Soil, Water and Environment, University of Dhaka. Soil was collected from Dhamrai in the district of Dhaka where groundwater contamination by arsenic has not been reported. The soil samples representing 0 - 15 cm depth from the surface were collected by composite soil sampling method as suggested by the Soil Survey Staff of the USDA.<sup>(8)</sup> The collected soil samples were made free from the visible roots and debris, air-dried, ground and sieved through a 0.5 mm stainless steel sieve. The sieved samples were then mixed thoroughly and used for further analysis.

Three different arum (*Colocasia antiquorum*) viz. Bilasi, Mukhi kachu-029 and Mukhi kachu-140 were procured from the Bangladesh Agricultural Research Institute (BARI), Gazipur. The treatments chosen were 0, 5, 20, 40 and 80 mg/l As applied through irrigation water. The background level of As in the collected soil sample was 1.5 mg/kg. This was considered as control and treatments were added in solution made with sodium meta-arsenite ( $\text{NaAsO}_2$ ). Four kg soil was put in pots of 4 L size having no holes at the bottom. The soil in each pot was mixed with urea, muriate of potash (MP) and triple super phosphate (TSP) as per requirement at rates estimated according to fertilizer recommendation guide.<sup>(9)</sup> Four arum seeds were sown in each of the pots and allowed to germinate. The pots were arranged in the net house in a completely randomized way. Positions of the pots were changed every alternate day to allow equal exposure of each of the pots to sunlight. Treatment plants received water with arsenic solution while the control plants received only tap water every alternate day from seed sowing till plant harvest. Each treatment was replicated thrice. The total amount of As treated irrigation water applied was recorded. Adequate plant protection measures were taken. Plants were harvested and uprooted at 90 days after seedling emergence. The harvested roots were washed with deionized distilled water several times to remove the soil adhering to it and to

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free the ions from the root free space. The aerial parts of plants were also washed. The plant samples were separated into three parts - root, leaf and stem. The collected plant samples were first air-dried and then oven dried at  $70 \pm 5^\circ\text{C}$  for 48 hours and the dry weight of plant sample was measured. The dried plant samples were then ground and sieved through a 0.2 mm sieve for further analysis. After harvesting, soil samples in each pot were also collected from the rhizosphere and prepared for analysis as mentioned above.

The various physical, chemical and physiochemical properties of the soils were determined following procedures described earlier.<sup>(10)</sup> Both plant and soil were analyzed for total arsenic by hydride generated atomic absorption spectrometry (HG-AAS) while iron and manganese were determined by AAS. Arsenic, iron and manganese of plant samples were extracted with  $\text{HNO}_3$  and of the soil with aqua regia solution.<sup>(11)</sup> Certified reference materials were carried through the digestion and analyzed as part of the quality assurance/quality control protocol. Reagent blanks and internal standards were used where appropriate to ensure accuracy and precision in the analysis of arsenic. Each batch of 20 samples was accompanied with reference standard samples to ensure strict QA/QC procedures.

### Results and Discussion

The collected soil samples were analyzed in the laboratory before the set up of the experiment to assess the nutrient status of the soil. Important soil properties are furnished as texture - silty clay, pH - 7.1, organic carbon - 0.91%, total N - 0.12%, total P - 0.02%, total K - 0.39%, available P - 2.69 mg/kg, available K - 3.14 mg/kg, aqua regia extractable As - 1.5 mg/kg, Fe - 0.15% and Mn - 550 mg/kg. Visual symptoms of the arums were keenly observed during the growth period. Plants of all the arums showed severe symptoms of As toxicity at rates of 40 and 80 mg/l As application. The reduced growth of plants and chlorotic spots on leaves confirmed the symptoms of As toxicity. The possibility of fungal disease symptoms (Late blight and Leaf spots) usually observed in arums could not be considered in this case as the control plants did not show any such chlorotic spots. The average dry weight of all the arums at different As treatments are presented in Fig. 1. The average dry weight of all the arums decreased with increasing arsenic concentration. Among the arums, "Bilasi" produced the highest amount of dry matter followed by "Mukhi-029" and "Mukhi-140". Two way ANOVA test was done for the dry matter production at different treatment levels among the arums. The results showed significant ( $p = 0.002$ ) negative effect of As treatment on dry matter production of plants as well as significant ( $p = 0.011$ ) difference among the arums. The results of the present investigation are in agreement with some earlier findings on other crops. Reduced growth due to As in growth medium has been reported for garden pea,<sup>(12)</sup> rice,<sup>(13, 14)</sup>

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wheat, lettuce and alfalfa<sup>(15)</sup> and for maize and tomato.<sup>(16)</sup> Reduced dry matter production was probably due to toxic effect of arsenic on physiological function of the plants. The detrimental effect of As in plants in the growth medium at higher concentrations might be due to its phytotoxic effect and its antagonism with the uptake of some other nutrient elements that might have suppressed plant growth and yield<sup>(12,13,15,16)</sup> or also could be due to susceptibility to disease because of ionic imbalance.

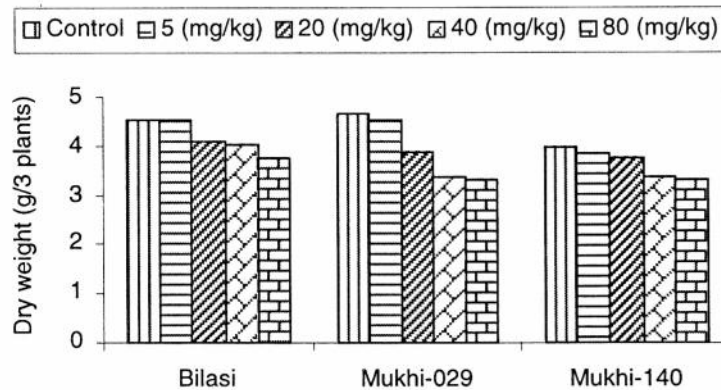


Fig. 1. Dry matter production of arums as affected by As treatment.

Arsenic concentration in roots, stems and leaves of all the arum samples were analyzed separately and results are shown in the Figs. 2, 3 and 4, respectively. Only mean values were used to compute the graphs.

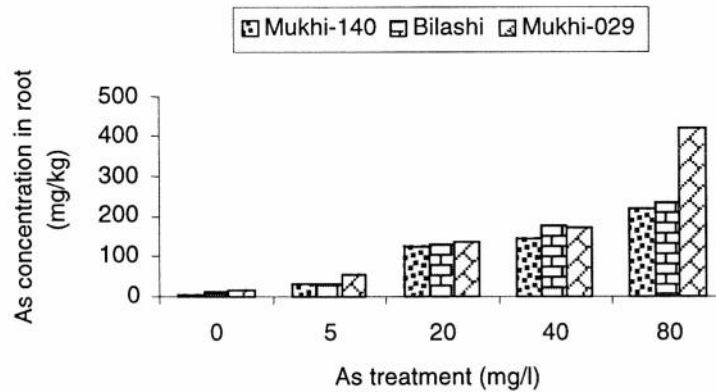


Fig. 2. Comparison of As concentrations in roots of all the arums.

The Fig. 2 shows that among the arum samples the concentration of As in roots was the maximum for Mukhi-029 followed by Bilashi while the least was for Mukhi-140 at different levels of As treatment. For stems and leaves, similar observations were made (Figs. 3 and 4). It was observed that, in general, As concentration in the

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plant parts increased with increasing rates of As treatments. However, at the highest rate of As (80 mg/l) application there has been a sudden drop in As concentration in

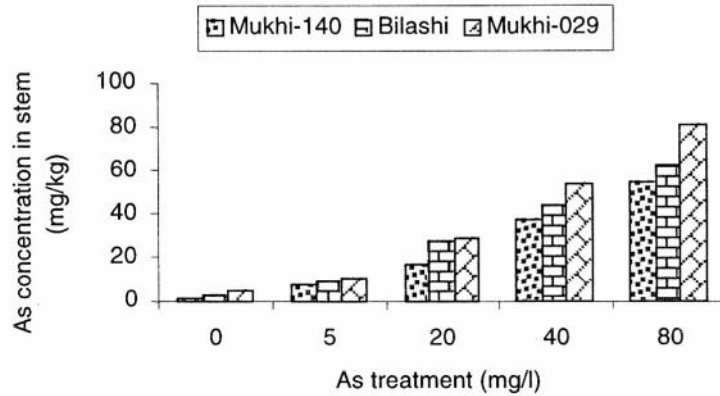


Fig. 3. Comparison of As concentrations in stems of all the arums.

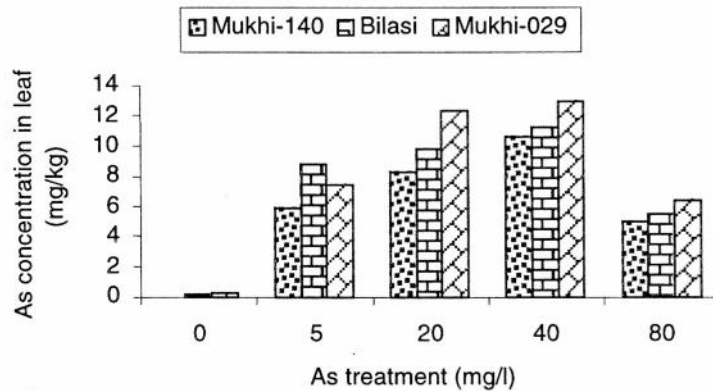


Fig. 4. Comparison of As concentrations in leaves of all the arums.

the leaves and the values were even lower than that of the observed for 5 mg/l As treatment. The maximum As concentration in roots were 417.85 mg/kg for Mukhi-029, 232.98 mg/kg for Bilashi and 218.95 mg/kg for Mukhi-140 at 80 mg/l As application. At 5 mg/l As application, no significant differences in arsenic concentration in either leaf or stem were observed. In general, the concentration of arsenic was found to be higher in roots followed by stems and leaves for any arsenic treatment. Higher accumulation of arsenic in roots of many plants has been reported; e.g., in Barmudagrass,<sup>(17)</sup> in Lodgepole pines<sup>(18)</sup> and in alfalfa.<sup>(19)</sup> This phenomenon has also been demonstrated by<sup>(20, 21)</sup> where they pointed out that the upward transport of As from roots was limited by its high toxicity to the radicular membranes of roots.

Fig. 5 shows the total As concentration in the whole plant of all the arum samples. It is clear from the graph that Mukhi-029 contained the maximum amount of arsenic followed by Mukhi-140 and Bilasi. It is also important to note that in control plants there were some As accumulation and that was perhaps due to the presence of water extractable As (native As) in soil. So, As concentration in arums followed the order: Mukhi-029 > Mukhi-140 > Bilasi and the plant parts followed the order: root > stem > leaf from maximum to minimum. The mean values of the experimental data were evaluated by employing linear-regression model of arsenic treatment against arsenic accumulation by edible parts of the three arums. The  $r^2$  values for Mukhi-029, Bilasi and Mukhi-140 were 0.996, 0.974 and 0.994, respectively indicating the fact that such high accumulation of As will invariably occur under similar situations.

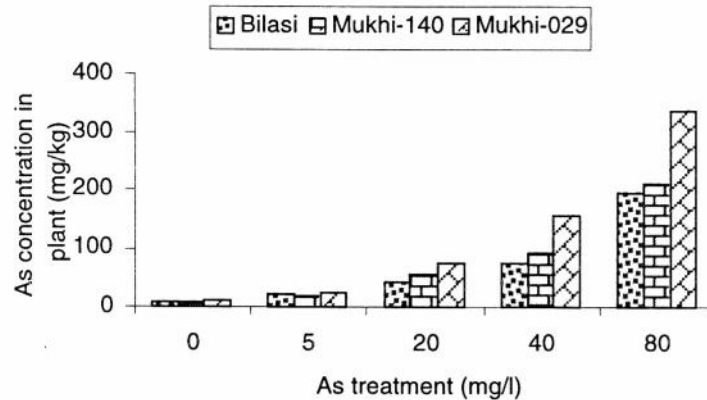


Fig. 5. Comparison of total arsenic concentration in whole plant of all the arums.

The mean values of As uptake (concentration  $\times$  dry matter produced) by the arums are presented in Table 1. The amount of As indicates the total amount applied to soil for the different treatments throughout the growing period. Amount of As in control indicates the initial soil As (1.50 mg/kg). The uptake of As showed an increasing order with the amount applied and this was shown irrespective of the arums. Among the different parts, As uptake in leaves was comparatively slower than either root or shoot. There was a decrease rather at 80 mg/l As application and that was probably due to decreased weight of leaf or physiological effect of As toxicity. Among the arums, Mukhi-029 accumulated the maximum amount of As in all its parts. The order of As uptake by leaves and roots of all the arum samples were: Mukhi-029 > Bilasi > Mukhi-140 and in stem the order was Mukhi-029 > Mukhi-140 > Bilasi from maximum to minimum. It may be due to the variation of dry weight of different parts of plants. Comparing the uptake by the whole plant the order followed as: Mukhi-029 > Mukhi-140 > Bilasi. At 80 mg/l As application, uptake of Mukhi-140

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Table 1. Arsenic uptake in arum.

| As spiked in soil (mg/kg) | Bilasi  |       |       |          |          | Mukhi-029 |       |       |          |          | Mukhi-140 |       |       |          |          |
|---------------------------|---------|-------|-------|----------|----------|-----------|-------|-------|----------|----------|-----------|-------|-------|----------|----------|
|                           | Root    | Stem  | Leaf  | T. plant | T. plant | Root      | Stem  | Leaf  | T. plant | T. plant | Root      | Stem  | Leaf  | T. plant | T. plant |
|                           | (mg/kg) |       |       |          |          | (mg/kg)   |       |       |          |          | (mg/kg)   |       |       |          |          |
| 1.5                       | 0.003   | 0.005 | 0.002 | 6.63     | 6.63     | 0.006     | 0.006 | 0.003 | 9.85     | 9.85     | 0.002     | 0.003 | 0.002 | 5.24     | 5.24     |
| 26.81                     | 0.021   | 0.005 | 0.003 | 19.62    | 19.62    | 0.025     | 0.007 | 0.004 | 23.81    | 23.81    | 0.017     | 0.004 | 0.003 | 18.63    | 18.63    |
| 102.75                    | 0.035   | 0.018 | 0.004 | 42.02    | 42.02    | 0.072     | 0.019 | 0.004 | 73.46    | 73.46    | 0.055     | 0.011 | 0.004 | 55.48    | 55.48    |
| 204                       | 0.073   | 0.022 | 0.005 | 73.5     | 73.5     | 0.145     | 0.023 | 0.005 | 155.98   | 155.98   | 0.080     | 0.020 | 0.005 | 92.91    | 92.91    |
| 406.5                     | 0.149   | 0.092 | 0.002 | 195.88   | 195.88   | 0.210     | 0.155 | 0.002 | 334.33   | 334.33   | 0.116     | 0.113 | 0.002 | 209.73   | 209.73   |



(209.73 mg/kg) and Bilasi (195.88 mg/kg) was very close. Two way ANOVA test was done and from the results it is clear that the effect of As treatment was highly significant ( $p < 0.001$ ) and the varietal difference was also significant at 0.06 level.

The transfer factors of all the arums in response to total As applied in soil are shown in the Table 2. Farrago and Mehra<sup>(22)</sup> have considered that when the transfer factor (plant/soil ratios) for any particular element is 0.1, then the plant can be considered as excluding the element from its tissues. In the present case, all the arums showed a transfer factor greater than 0.1 meaning that arums in general have a great affinity to accumulate As from the growth medium. The results further confirm the earlier findings by Huq and Naidu<sup>(4)</sup> that arums, in general, are As accumulators. A very high transfer factor for control plants further substantiates this observation. It could thus be concluded that As extraction by arums are quite significant. Among the arums, transfer factor was the maximum for Mukhi-029, signifying the fact that this material has the greatest affinity for As uptake from soil in all its parts.

**Table 2. Transfer factor of arsenic of all the arums.**

| Total As in soil<br>(mg/kg) | Transfer factor |           |           |
|-----------------------------|-----------------|-----------|-----------|
|                             | Bilasi          | Mukhi-029 | Mukhi-140 |
| 1.50                        | 4.42            | 6.57      | 3.50      |
| 26.81                       | 0.73            | 0.89      | 0.69      |
| 102.75                      | 0.41            | 0.71      | 0.54      |
| 204.00                      | 0.36            | 0.76      | 0.46      |
| 406.50                      | 0.48            | 0.82      | 0.52      |

**Table 3. Correlation coefficient of plant P, Mn and Fe content with arsenic content for all the arums.**

| Arum      | As vs P |         | As vs Mn |         | As vs Fe |         |
|-----------|---------|---------|----------|---------|----------|---------|
|           | r value | p value | r value  | p value | r value  | p value |
| Bilasi    | 0.478   | 0.072   | 0.106    | 0.707   | 0.395    | 0.145   |
| Mukhi-029 | 0.574   | 0.025   | 0.192    | 0.492   | 0.319    | 0.247   |
| Mukhi-140 | 0.297   | 0.283   | 0.058    | 0.838   | 0.307    | 0.266   |

Increased As concentration had a pronounced effect on the nutrient uptake as well as transport in plants. The effect of As on P, Fe and Mn contents in arums was evaluated. Plant phosphorus concentration increased with increasing As concentrations. Pearsons correlation coefficient and p value of As (mg/kg) and P (%) content in



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all the arums are presented in Table 3. A noticeable positive correlation was observed for all the arums but the correlation was highly significant for only Mukhi-029. Such a positive correlation could be due to the fact that arsenic is chemically similar to phosphorus<sup>(21)</sup> and roots take up arsenic and phosphorus by the same mechanism.<sup>(23)</sup> The correlation coefficient and p values between arsenic and manganese as well as arsenic and iron content of all arums are also presented in Table 3. A positive but statistically insignificant correlation was observed for all the arums in both the cases.

From the present findings it is clear that arsenic accumulation in the arums differed significantly from one another. Among the arums, Mukhi-029 accumulated the maximum amount of arsenic compared to either Mukhi-140 or Bilasi. The literature reports that arum (*Colocasia antiquorum*) showed very high accumulation of arsenic (153.2 mg/kg dry wt.) grown with As contaminated water.<sup>(4)</sup> These authors studied the accumulation of As in different parts of arum, though nothing was done on the response of different arum. Very high level of arsenic in taro (loti from arum plant, 440 µg/kg) has also been reported.<sup>(24)</sup> In a more recent study,<sup>(25)</sup> the leaf of arum was found to contain between 90 and 3990 µg/kg of arsenic. In the present study it was clear that As uptake by root was the maximum for the arum Mukhi-029 (334.33 mg/kg) compared to Mukhi-140 (209.73 mg/kg) and Bilasi (195.88 mg/kg). It could thus be concluded that, Bilasi could preferably be grown in As contaminated areas to minimize As ingestion through arum.

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