



## INFLUENCE OF MANGANESE AND PHOSPHATE ON MITIGATION OF ARSENIC TOXICITY IN CHILLI (*Capsicum annum* L.)

Dipak Girhepuje and P.K. Bandopadhyay

Department of Plant Physiology, Bidhan Chandra Krishi Viswavidyalaya, Mohanpur, Nadia, West Bengal (India)-741252

Corresponding author, E-mail: [dipakgirhepuje@gmail.com](mailto:dipakgirhepuje@gmail.com)

### ABSTRACT

The nature of arsenic toxicity in chilli seedlings (variety-Bulleete) were studied at Bidhan Chandra Krishi Viswavidyalaya during the year 2010-11. All treatments were given at the same time as soil application to plants after 20 days, 50 days and 80 days of transplanting. The supply of arsenate (5 mg As/Kg soil) did not show toxic effect at the stage of 30 days growth but at later stages of growth, arsenate was found toxic on fresh fruit weight, dry fruit weight and fresh test weight. The inhibitory effect of arsenate was less pronounced at the beginning but increased with time. It decreased starch and total carbohydrate content with the decline in the later stages of growth at high concentrations of arsenate. The combined application of arsenate with manganese (1 ppm/pot), arsenate with phosphate (13 mg/Kg soil) and arsenate, manganese, phosphate together exhibited significant beneficial alterations of all the parameters tested under the purview of arsenate treatment alone. It was observed a better increment in fruit weight and biosynthesis of carbohydrate with the application of manganese and phosphate in chilli seedlings.

**Key Words :** Manganese (Mn), phosphate (P), arsenic (As), amelioration, chilli

Chilli (*Capsicum annum* L. and *Capsicum frutescens* L.) is the universal spice of India belonging to the family-Solanaceae. Chilli is one of the most valuable crops of India. The crop is grown largely for its fruits all over the India. It occupies an important place in Indian dietary. It is used as a principle ingredient of various curries and chutneys. It is also used for vegetables, spices, condiments, sauces and pickles. Dry chillies are used for curry powder. Commercial part i.e. green as well as ripe and dried pod (fruit) is essential ingredient of human consumption in every day. Chilli, a native of tropical zone is widely cultivated for use as spices and vegetables in temperate zones as well as throughout tropics. Green chilli is rich in vitamin-A, thiamine, riboflavin, nicotinic acid and also minerals P, K and Ca. Pungency of chilli is due to an alkaloid, Capsaicin ( $C_{18}H_{27}NO_3$ ) and the red colouration is due to the pigment Capsanthin.

The productivity of the crop generally fluctuates with the instability of environmental factors and in many cases decreases the yield and quality of the crops. There is another factor, underground irrigation of land which is generally man made pollutes the surface soils by arsenic. Arsenic is an element that is non essential and toxic to plants. Its accumulation in food crops may pose a health risk to humans. It is metalloid compound found in arsenite and arsenate form and very much

toxic to animal and plant system when mixed in metabolic pathways. Among the two forms available in soil, arsenite is highly toxic to plant system, which ultimately drastically hampers the growth of the plant. The main affected part is the root system and ultimately damages to the shoot system. These arsenite/arsenate when in tissue system in substantial amount, decline the productivity of the crop and it is very difficult to get rid of. To overcome this problem, simple economical physiological treatments were chosen to detoxify the toxicity effect of arsenic by applying manganese and phosphate in very small concentration in different growth phases of the plants. The beneficial effects of treatments were analyzed through physiological and biochemical parameters and observed values at least gave certain betterment effect over arsenate treatment, which can be recommended to our farmers to conquer the problem.

### MATERIALS AND METHODS

The experiment was carried out with 4 replications at University experimental farm and also in laboratory condition, BCKV, Mohanpur, Nadia, West Bengal, India during the period of 2010 and 2011. The seedlings of *Capsicum annum* (variety-Bulleete) were collected from Bidhan Chandra Krishi Viswavidyalaya (BCKV) farm, Mohanpur and planted in earthen pots of 10 inch diameter (5 plants per pot) for future study. Before

transplanting the plants, pots were prepared by pouring 5 kg soil in each pot, the soil was added thoroughly with standard doses of fertilizers (N: K @ 90: 60 kg/ha) and organic manures fortified into the soil. The solutions of sodium arsenate and manganese chloride were prepared by dissolving it in the distilled water. Quantity of salt is taken in such a way that it would contain 5 mg As/kg soil in a pot, 1 ppm Mn/pot and Phosphate (P) - 13 mg/kg soil for each time. Solution was prepared properly just before application.

**Application of treatments :** Treatments which were given as soil application to plants after 20 days, 50 days and 80 days of transplanting are as follows :

- Arsenic as sodium arsenate as IV ( $\text{Na}_2\text{HAsO}_4 \cdot 7\text{H}_2\text{O}$ ) 21.4235 mg/kg soil (5 mg As/kg soil)
- Manganese as manganese chloride ( $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ ) – 3.6 mg manganese chloride salt in 1000 ml of distilled water (1 p.p.m.)/pot
- Phosphate (P) - 13 mg/kg soil

**Recording the Data :** The individual data recorded during different growth phases of plant at 30 DAT (DAT-days after transplanting), 60 DAT, 90 DAT, 80 DAT, and 100 DAT by destructive sampling method and were considered separately for statistical analysis which was done by RBD (Randomized Block Design) method. The parameters considered for analysis may be grouped in the following heads :

- **Physiological parameters:** Fresh fruit weight, Dry fruit weight, Fresh test weight.
- **Biochemical parameters :** Starch, Total carbohydrate.

Starch was estimated by anthrone reagent. Sample was extracted with hot 80% ethanol to remove sugars till the washings do not give colour with anthrone reagent. Residue was extracted at 00C with fresh perchloric acid and supernatant was collected. Anthrone reagent was added in each tube containing supernatant. Tubes were heated followed by cooled and read out the intensity of green to dark green colour at 630 nm. From the standard graph drawn, the contents of starch were calculated and expressed in terms of percentage (1).

Total carbohydrate was determined according to the method given by (2). Finely cut leaves/fruits samples were hydrolyzed with 2.5 N HCl and the

**Table-1 :** Toxicity effect of Arsenate (As IV) and its amelioration by Manganese and phosphate on fresh and dry fruit weight.

| Parameters | Fresh fruit weight per fruit (gm) |         | Dry fruit weight per fruit (gm) |         |
|------------|-----------------------------------|---------|---------------------------------|---------|
|            | 80 DAT                            | 100 DAT | 80 DAT                          | 100 DAT |
| Control    | 7.49                              | 8.10    | 1.46                            | 1.81    |
| AS         | 7.07                              | 7.24    | 1.18                            | 1.50    |
| As+Mn      | 7.68                              | 8.11    | 1.51                            | 1.86    |
| As+P       | 7.80                              | 8.70    | 1.83                            | 2.24    |
| As+Mn+P    | 8.28                              | 9.35    | 2.01                            | 2.58    |
| C.D. at 5% | 0.69                              | 0.95    | 0.35                            | 0.37    |
| S. E.      | 0.21                              | 0.29    | 0.11                            | 0.11    |

**Table-2 :** Toxicity effect of Arsenate (As IV) and its amelioration by manganese and phosphate on fresh test weight.

| Parameter  | Fresh Test Weight (gm) |         |
|------------|------------------------|---------|
|            | 80 DAT                 | 100 DAT |
| Control    | 22.33                  | 33.64   |
| AS         | 20.61                  | 30.97   |
| As+Mn      | 24.51                  | 36.13   |
| As+P       | 27.71                  | 46.38   |
| As+Mn+P    | 30.24                  | 49.54   |
| C.D. at 5% | 3.74                   | 4.56    |
| S. E.      | 1.15                   | 1.40    |

**Table-3 :** Starch content from the leaves and fruits of chilli plants under different growth phases treated with arsenate and ameliorated by manganese and phosphate.

| Parameters | Starch (%) |        |       |
|------------|------------|--------|-------|
|            | 30 DAT     | 90 DAT | Fruit |
| Control    | 2.44       | 2.67   | 0.18  |
| AS         | 3.19       | 1.52   | 0.09  |
| As+Mn      | 3.16       | 3.17   | 0.12  |
| As+P       | 3.30       | 2.63   | 0.18  |
| As+Mn+P    | 3.47       | 3.10   | 0.23  |
| C.D. at 5% | 0.59       | N.S.   | 0.08  |
| S.E.       | 0.19       | 0.49   | 0.03  |

**Table-4 :** Total carbohydrate content from the leaves and fruits of chilli plants under different growth phases treated with arsenate and ameliorated by Manganese and Phosphate.

| Parameter  | Total Carbohydrate (%) |        |       |
|------------|------------------------|--------|-------|
|            | 30 DAT                 | 90 DAT | Fruit |
| Control    | 3.37                   | 4.55   | 0.96  |
| AS         | 4.87                   | 2.51   | 0.67  |
| As+Mn      | 5.00                   | 4.33   | 1.60  |
| As+P       | 4.92                   | 3.90   | 0.97  |
| As+Mn+P    | 4.68                   | 4.73   | 1.34  |
| C.D. at 5% | 0.53                   | N.S.   | N.S.  |
| S.E.       | 0.18                   | 0.53   | 0.22  |

supernatants were used for sugar estimation. 4 ml anthrone reagent was added to all the tubes containing supernatant and heated the tubes for 8 minutes. After

cooling the tubes, read out the intensity of green to dark green colour at 630 nm. From the standard graph carbohydrate contents of sample were calculated and represented in terms of percentage.

## RESULTS AND DISCUSSION

In respect of productivity of the chilli crop fresh fruit weight per fruit and dry fruit weight per fruit were recorded to compare the treatment effect with As and control which are presented in table-1. It was observed from the result that in two plucking days such as 80 days and 100 days, the fresh and dry fruit weight per fruit decreased with As treatment but ameliorating treatments showed significant increase in fruit weight. The maximum fruit weights were reported with As+Mn+P treatments in both the plucking stages. Both the parameters studied, increased in the ameliorating treatments over the As possibly due to better absorption of Mn and P through root system and translocated to the upper region of the plant and on the contrary there was a less absorption of As by root.

Fresh test weights under 80 and 100 days after transplanting were recorded for strong evidence of the study among the different treatments which are presented in table 2. It was observed from the result that in two plucking days, fresh test weights decreased with As treatment but ameliorating treatments showed significant increase in test weights. The maximum test weights were reported with As+Mn+P treatments in both the plucking stages. It had happened because arsenic toxicity affected photosynthesis which ultimately resulted in the reduction of test weight (3).

In table-3, a starch concentration in different treatments has been presented. There was no much difference in starch accumulation in As treatment under different stages of growth and fruits compare to control one. When As contaminated plant treated with Mn or P or both, there was accumulation of starch in early growth phases of plants but very marginal accumulation in late stage of seedling and fruits of chilli. It was reported in the earlier literature that As toxicity disturbances the carbohydrate metabolism and decreases starch content during the last part of the growing period (4). Also it was observed in rice that due to As exposure there was an increase in

starch content and this may be due to activity of starch phosphorylase and on the contrary  $\alpha$ -amylase activity decreases (5).

In table-4, total carbohydrate content in different growth phases has been presented. While analyzing the total carbohydrate content it was found that initially there was an increase in total carbohydrate content in all the ameliorating treatments along with As up to 30 days of growth compared to control set. The prominent decline in total carbohydrate observed in 90 days old seedlings as well as fruits of chilli. The total carbohydrate content in all the cases showed negative results. Arsenate also affected the activities of different enzymes of carbohydrate metabolism. The present experimental studies with total carbohydrate to some extent restore with ameliorating treatment compared to As. On the whole it showed marginal improvement over As treatment (6).

From the overall results and discussion we may conclude that As under moderate to high doses caused serious physiological and biochemical disturbances in plant system and that could be minimised by the application of Mn and P in proper doses.

## REFERENCES

1. Thayumanavan, B. and Sadasivam, S. (1984) : Estimation of starch by anthrone reagent. *Qual. Plant Foods Hum. Nutr.* 34 : 253-259.
2. Hodge, J. E. and Hofreiter, B. T. (1962): Determination of total carbohydrates by anthrone method. *Methods Carbohydr Chem* 1 : 380.
3. Rahman, M.A.; Hasegawa, H.; Rahman, M.M.; Islam, M. N.; Miah, M.A.M. and Tasmen, A. (2007): Effect of arsenic on photosynthesis, growth and yield of five widely cultivated rice (*Oryza sativa* L.) varieties in Bangladesh. *Chemosphere*. 67(6) : 1072-1079.
4. Chang SiMin, Ma XinMing, Zhang GuiLong, Xiong ShuPing, Zhan KeHui and Liu GuoShun (2006): Effects of arsenic toxicity on carbon and nitrogen metabolism and the yield and quality of flue-cured tobacco. *Journal of Plant Ecology*. 30 (4) : 682-688.
5. Bhaskar Choudhury, Souvik Mitra and Biswas, A. K. (2010) : Regulation of sugar metabolism in rice (*Oryza sativa* L.). Seedlings under arsenate toxicity and its improvement by phosphate. *Physiology and Molecular Biology of Plants*. 16(1) : 59-68.
6. Jha, A. B. and Dubey, R. S. (2004): Carbohydrate metabolism in growing rice seedlings under arsenic toxicity. *Journal of Plant Physiology*. 161(7) : 867-872.