



RESEARCH ARTICLE

Morphology & Productivity Response of *Phaseolus aconitifolium* under the Stress of Lead Nitrate [Pb(NO₃)₂]

Astik Kumar Buts and Vijaya L. Chaudhary

P.G. Dept. of Botany, D.S. College, Aligarh, U.P., India

Email: dr.astikbut1968@gmail.com, vijayaa_110@rediffmail.com

Received: 12th May 2015, Revised: 19th June 2015, Accepted: 25th June 2015

ABSTRACT

The present experiment was conducted to evaluate the morphological & productivity response on *Phaseolus aconitifolium* (Moth Bean) under the stress of lead nitrate. The seeds were treated with 0.1%, 0.25%, 0.50% & 1% concentration of [Pb(NO₃)₂] before sowing in the experimental plots. The seeds obtained from M₁ generation were further treated with corresponding lead nitrate concentration before sowing in experimental plots to obtain M₂ generation. We observed that lead nitrate has very deleterious effect on morphology & productivity of *Phaseolus aconitifolium*. It showed a decreasing trend with increasing treatment concentration of lead nitrate. Reduction among morphological features was maximum in 01% treatment concentration. Germination, plant height, no. of branches reduced up to 37.1% while no. of pods per plant reduced 45.07% & weight of seeds reduced up to 29.85% in M₁ generation. But these all slightly recovered in M₂ generation which shows development of tolerance towards the lead nitrate stress in successive generations. It may be due to occurrence in changes in normal physiological functions and biochemical synthesis under stress condition of metal toxicity.

Key words: *Phaseolus aconitifolium*, [Pb(NO₃)₂], Morphology, Productivity, Generation

INTRODUCTION

Moth bean is one of the important legume widely grown in arid and semi-arid parts of the country. It is a minor kharif pulse crop and considered as one of the most drought tolerant among the grain legumes. The drought tolerance of this bean due to well-developed tap root system or the manner in which the plant produce a thick low lying mat covering the soil surface and apparently reducing moisture loss.

Moth bean is a good source of protein (24%) and rich in dietary fibers. In India, it is grown in an area of 1.65 million hectares, mostly confined to Karnataka, Gujarat, Rajasthan, Maharashtra and Haryana with a productivity of 0.48 million tons. This crop is cultivated alone and in various cropping systems on plains and sand dunes. Its water requirement is very low. On average the crop transpiration rate is 18-22 mm per day during emergence and early growth stage of crop, with a maximum of 4.8 mm per day during flowering and pod formation stages (Singh *et al.*, 2000). It is highly susceptible to yellow mosaic virus disease.

Lead is found in the environment in various forms. Today, most large scale heavy metals are the common types (Nriagu, 1992). Lead contamination is an important problem especially in agriculturally developed zone. The contaminants around industrial areas are a serious problem. Such contaminants is largely injurious due to anthropogenic activities such as indiscriminate use of pesticides, containing heavy metals in agriculture, discharge of untreated industrial waste disposal, high rate of burning of fossil fuels, mining etc. As an indicator of phytotoxicity, various authors have reported time to time different bio-monitoring indices based on germination and seedling growth to indices metal stress effects using different plant systems (Baki and Anderson, 1973; Mahatre and Chaphekar, 1982; Buts *et al.*, 2013).

Soil contaminated with Pb causes sharp decrease in crop productivity thereby posing a serious problem for agriculture (Johanson and Eaton; 1984). Although Pb is not essential nutrients for plants but majority of lead is easily taken up by plants from soil and accumulated in root while,

only a small fraction was translocated upwards to the shoots (Patra *et al.*, 2004). Pb affects several metabolic activities at different cell compartments. The effect of Pb depends on concentration, type of soil, soil properties and plant species. Pb toxicity leads to decrease germination percent, length and dry mass of root and shoot (Mouzuroglu and Geckil, 2002), distributed mineral nutrition (Pavivoke, 2002), reduction in cell division (Eun *et al.*).

According to Shafiq *et al.* (2008) decrease in seed germination of plant can be attributed to the accelerated breakdown of store material in seed by the application of heavy metal mixture. Dalal and Bairgi (1985) have found reduction in seed germination, root, shoot and seedling length of jute varieties, *Corchoru solitorius* JRO 524 and *Capsular corchorus* JRC 321 at different levels of Pb, particularly at 20 mg/l. Haghiri (1973) reported that the toxicity of some metal is so high that plant growth is retarded before a large quantity of an element can be translocated.

Many workers have reported the effect of metal toxicity on different plants taking few character under study. But none have studied the effect of metal toxicity on whole life cycle of the plant. The purpose of this study is to asses the effect of lead nitrate on morphology and productivity of *Phaseolus aconitifolium* up to two generation.

MATERIALS AND METHOD

PLANT MATERIAL:

The plant material involved in the present investigation was *Phaseolus aconitifolium* (Moth bean) with $2n=22$. It belongs to family Fabaceae. The seeds of Moth bean were obtained from Central Arid Zone Agricultural Institute, Jodhpur. Healthy seeds of equal size & shape were selected for treatment with lead nitrate.

Chemical used for investigation: Lead Nitrate [$Pb(NO_3)_2$] is taken for experiment in vivo study of *Phaseolus aconitifolium*. The treatment concentrations taken for study are 0.1%, 0.25%, 0.50% & 01%.

MORPHOLOGICAL & REPRODUCTIVE STUDIES IN FIELD:

Parameters taken for the study of the impact of different concentrations (0.1%,0.25%,0.50% & 01%) of lead nitrate were germination, height of plants, number of branches, time taken for first flowering, number of pods per plant, weight of 100 seeds, period of harvesting etc. Prior to the experiments, healthy seeds of equal size & shape were selected. 100 seeds were used for each treatment.

Before sowing in the field, seeds were soaked in water for 4 hours. Then soaked 100 seeds were placed in each petridishes containing concentration of 0.1%, 0.25%, 0.50% & 01% Lead nitrate for 2 hours in the laboratory and then the seeds were sown in the experimental plots (field) under controlled & protected conditions. 100 seeds soaked in water for six hours sown in experimental plot as control. The experiments were set in 5 blocks. The seeds were sown in lines keeping distance of 15 cm. between the plants and 25 cm. between the lines.

In the field the emergence of hypocotyls and cotyledon above the surface of the soil had been taken as an index of germination. Arrangements were made for regular weeding and irrigation. Neither chemical nor other fertilizers were used. This was done to avoid confusion. Seeds of moth bean were sown in the first week of July and harvesting was done in the 2nd week of October. In between the above periods morphological & reproductive characters were studied with respect to plant height, no. of branches per plant, date of initiation of flowering, period of harvesting, number of pods per plant etc. Height of the plant was recorded at the time of maturity. After harvesting, weight of hundred seeds was recorded from control as well as from the treated plants. This was considered as M_1 generation.

Mature seeds of M_1 generation from the plants treated with different concentrations were harvested separately and stored separately. These seeds were used next year in the same way after giving treatment with corresponding concentrations of lead nitrate taken for study and resulted crop was considered as M_2 generation. The seeds of each set were treated with corresponding concentration of lead nitrate. Morphological characters were recorded in M_1 & M_2 generations and finally the phenotypic variability and pod productivity were calculated. Raw

data collected is compiled by standard method. We calculated the mean of the observed data and find out the standard deviation to draw conclusion.

RESULT AND DISCUSSION

The results obtained in present experiment have been shown in table 1 & 2 and figure 1, 2, 3, 4 & 5 and expressed together with the discussion in separate heading as under :-

Table 1: Effect of Lead nitrate on *Phaseolus aconitifolium* in M₁ generation

Treatments (Lead nitrate)	Germination of seeds in the field	Height (cm)±SD	No. of branches per plant ±SD	Days taken for 1 st flowering	Period of harvesting (In Days)	No. of pods/plant±SD	Weight of 100 seeds (gram)±SD
0.1%	82%	13.3±1.7	3.1±1.6	27-37	62-65	24.0±1.3	1.420±0.18
0.25	76%	13.1±1.6	3.1±1.7	27-37	62-65	23.5±2.9	1.363±0.31
0.50%	70%	12.7±1.3	3.0±1.6	26-36	62-65	21.0±3.1	1.346±0.78
01%	60%	12.2±0.2	2.8±1.5	26-36	62-65	19.5±1.2	1.304±0.56
Control	93%	19.4±1.6	3.7±1.8	28-38	62-65	35.5±1.4	1.859±1.41

Table 2: Effect of Lead nitrate on *Phaseolus aconitifolium* in M₂ generation

Treatments (Lead nitrate)	Germination of seeds in the field	Height (cm)±SD	No. of branches per plant ±SD	Days taken for 1 st flowering	Period of harvesting (In Days)	No. of pods/plant±SD	Weight of 100 seeds (gram)±SD
0.1%	85%	18.8±2.5	3.0±1.7	25-35	60-65	52.6±3.6	2.241±1.6
0.25	80%	18.0±2.4	2.9±1.70	25-35	60-65	48.6±4.6	2.046±0.6
0.50%	72%	17.9±1.3	2.9±1.70	21-31	60-65	44.3±3.3	2.037±1.4
01%	65%	16.9±1.8	2.7±1.64	21-31	60-65	42.9±2.6	1.926±2.2
Control	100%	22.3±1.94	3.8±1.94	25-35	60-65	51.85±3.1	2.197±0.16

EFFECT ON SEED GERMINATION

The germination percentage in M₁ generation was found to be 82%, 76%, 70% and 60% in 0.1%, 0.25%, 0.50% and 01% treatment concentration of lead nitrate respectively. The percentage germination in M₂ generation was 85%, 80%, 72% and 65% in 0.1%, 0.25%, 0.50% and 01% treatment concentration of lead nitrate. In control, it was 93% in M₁ generation and 96% in M₂ generation.

The germination percentage was decreased when the lead concentration increased, which shows that higher lead concentration inhibit germination (Fargasova, 1994; Kiran and Munzuroglu, 2004; Wierzbicka and Obidzinska, 1998). This can also be due to toxic effects of ions on the germination process (Khajeh-Hosseini *et al.*, 2003).

EFFECT ON PLANT HEIGHT

The average heights of *Phaseolus aconitifolium* were observed. The observed heights were 13.3 cm, 13.1 cm, 12.7 & 12.2 cm in 0.1%, 0.25%, 0.50% & 01% concentration of lead nitrate treated seeds respectively in M₁ generation while these are 18.8 cm, 18.0, 17.9 & 16.9 cm in 0.1%, 0.25%, 0.50% & 01% concentration of lead nitrate treated seeds respectively in M₂ generation. The average height of the plants was 19.4 cm & 22.3 cm in M₁ & M₂ generations respectively under control. Thus it is clear that with increase in the concentration of lead nitrate for treatment, the height of the plants decreases respectively.

The decrease percentages of height of the plants are 31.5%, 32.5%, 34.5% & 37.1 in 0.1%, 0.25%, 0.50% & 01% concentrations of lead nitrate treated seeds respectively in M₁ generation while, it were 15.7%, 19.4%, 20.2% & 24.21% in M₂ generation. Therefore, it is enunciated that the percentage decrease in the height of the plants increases with increase in the treatment concentration of lead nitrate. But the decrease percentage is higher in M₁ generation in comparison to the M₂ generation under the stress of same concentration of lead nitrate. It shows

that the seeds have developed adaptability up to some extent in M₂ generation towards each treatment solution.

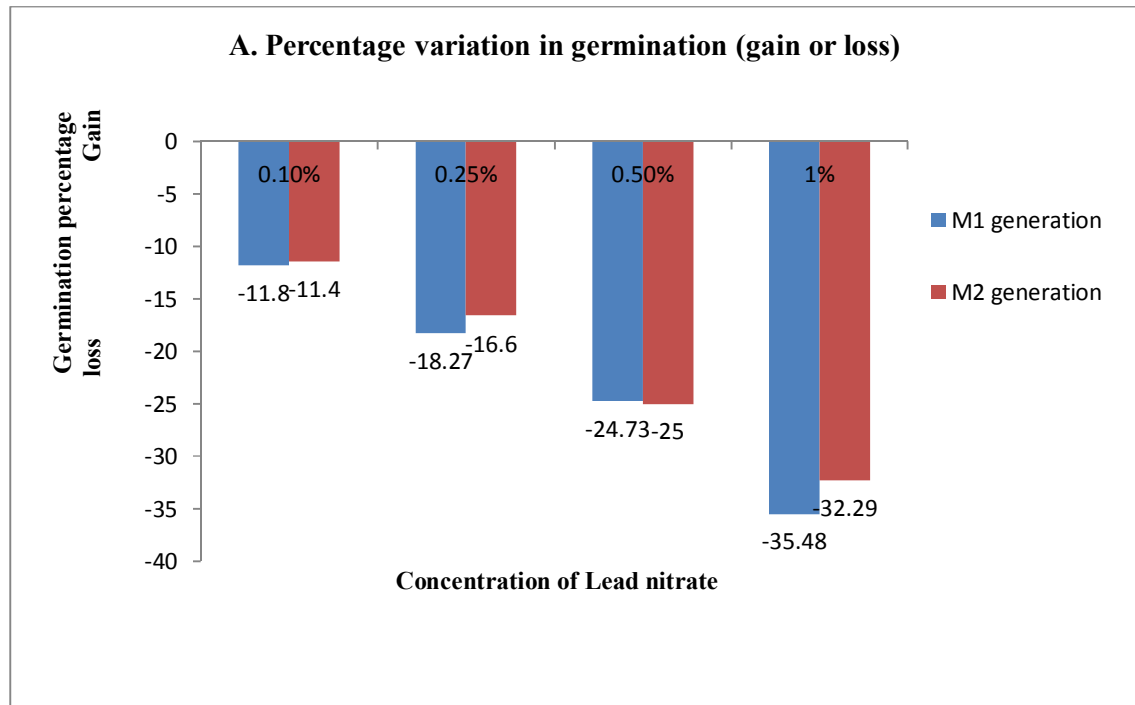


Fig. 1: Percentage variation in germination of *Phaseolus aconitifolium* treated with Lead nitrate in respect to control

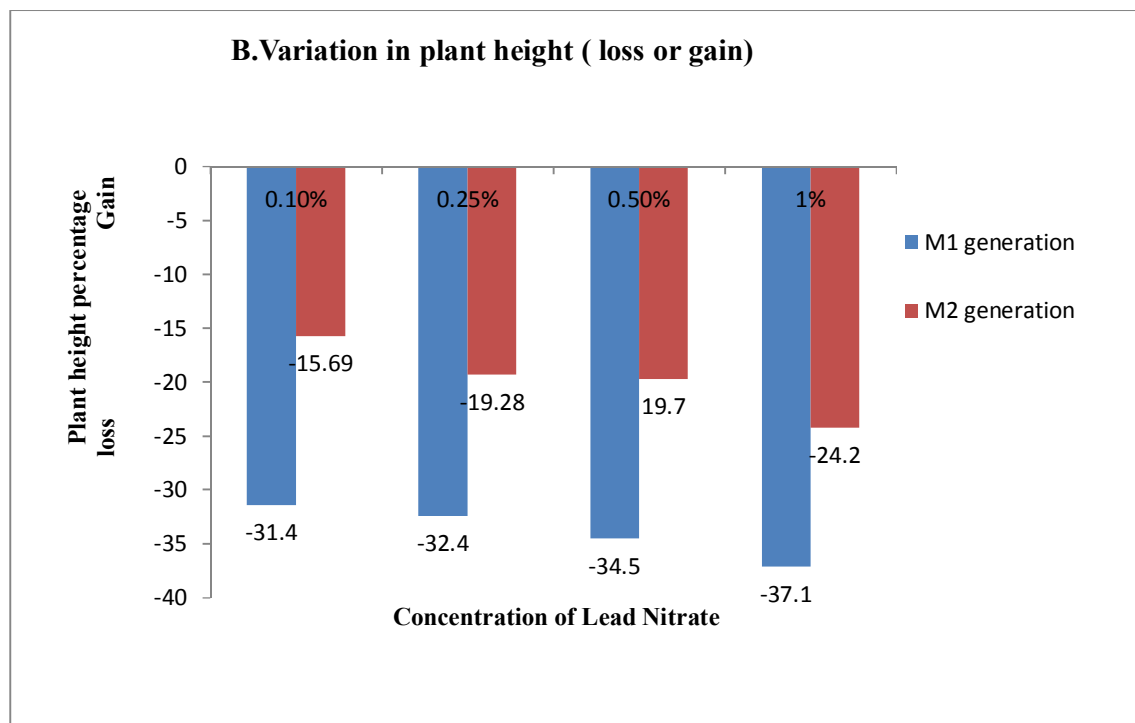


Fig. 2: Percentage variation in plant height of *Phaseolus aconitifolium* treated with Lead nitrate in respect to control

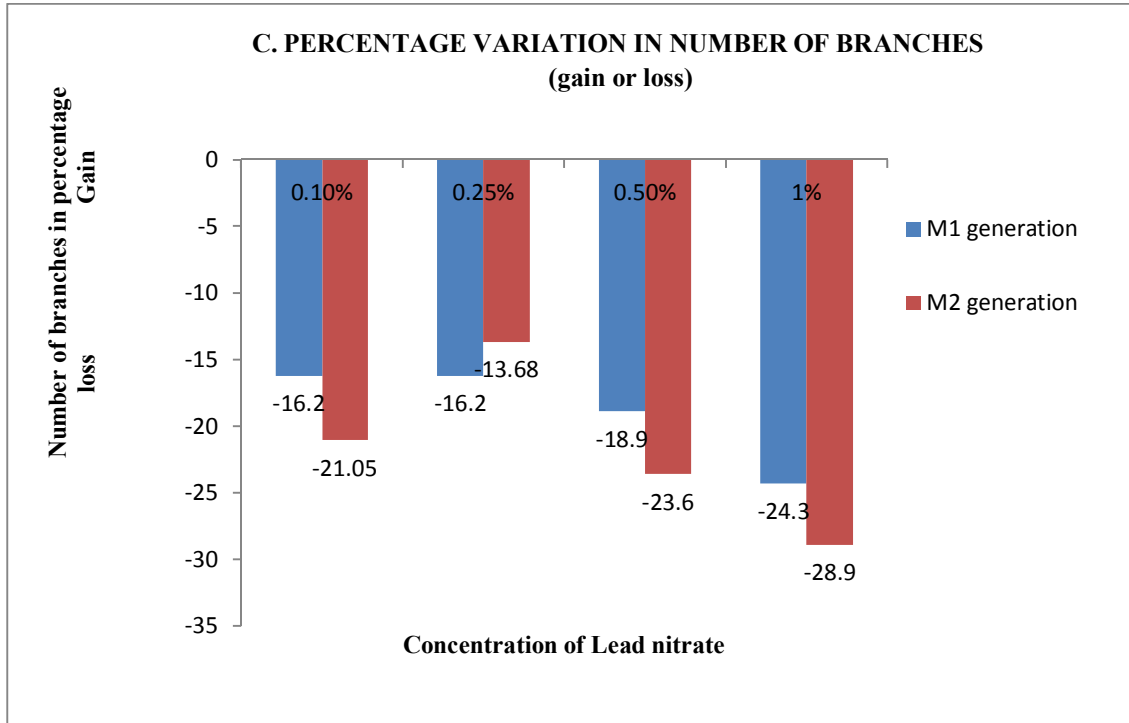


Fig. 3: Percentage variation in average no. of branches of *Phaseolus aconitifolium* treated with Lead nitrate in respect to control

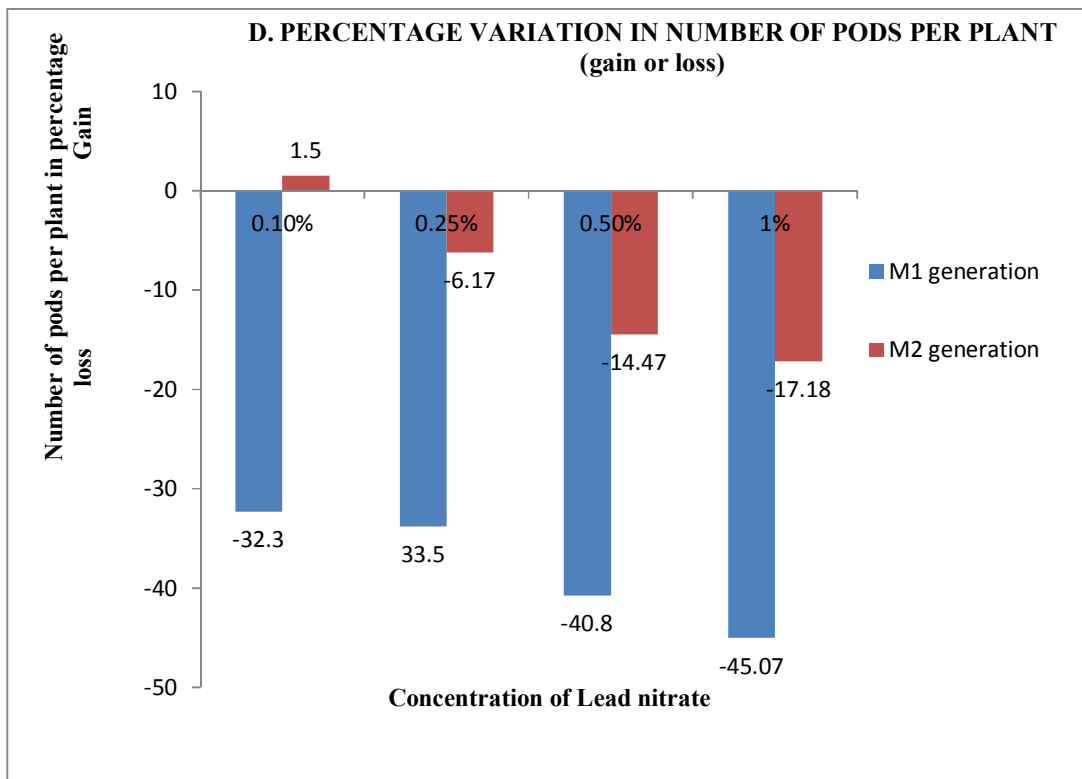


Fig. 4: Percentage variation in average number of pods per plant of *Phaseolus aconitifolium* treated with Lead nitrate in respect to control

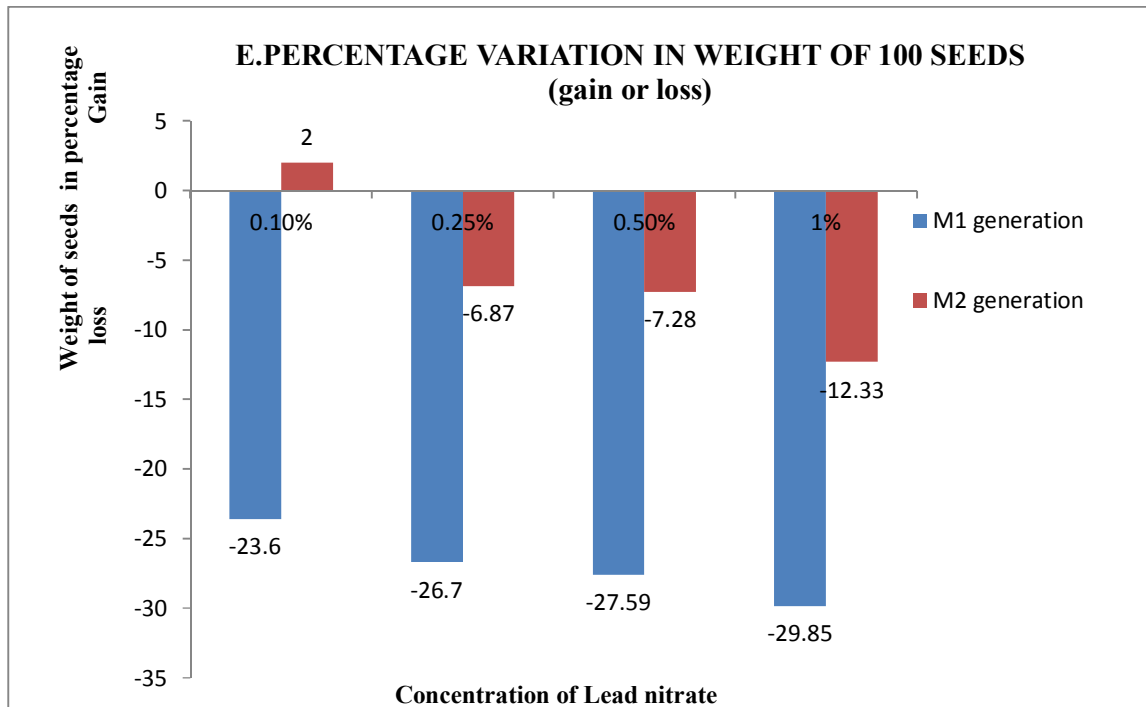


Fig. 5: Percentage variation in weight of seeds of *Phaseolus aconitifolium* treated with Lead nitrate in respect to control

Reduction in plant height could be attributed to the adverse effect of lead nitrate treatments on cell elongation and cell expansion. The effects of toxic substances on plants are dependent on the amount of toxic substances taken up from a given environment. The other reason of reduced height of the moth bean with increasing concentrations of metal treatments could be due to the reduction in meristematic cells present in this region and some enzymes contained in the cotyledon and endosperm. Cells become active and begin to digest and store food which is converted into soluble form and transported to the radical and plumule tips e.g. enzyme amylase convert starch into sugar and protease act on protein. So, When enzymatic activities were affected, the food did not reach to the radical and plumule and in this way shoot and seedling length were affected (Kabir *et al.*, 2008).

EFFECT ON NUMBER OF BRANCHES

Number of branches were 3.1, 3.1, 3.0 and 2.8 in M₁ generation in treatment concentration of 0.1%, 0.25%, 0.50% and 01% of lead nitrate respectively while 3.7 branches was recorded in control. In M₂ generation, average numbers of branching were 3.0, 2.9, 2.9 and 2.7 while it was 3.8 in control. It is decreasing with increasing concentration of lead nitrate.

Effect is deductive on *Phaseolus aconitifolium* regarding the branching with increasing treatment concentration of lead nitrate. The *Phaseolus aconitifolium* is a pulse of arid zone which is different from the plants of the other region. It can be also concluded from the observation that tolerance power towards lead nitrate of the plants of arid zone is less in comparison to the plants of temperate zone. However, what type of biochemical process is responsible for this difference is a matter of further research.

DAYS TAKEN FOR FLOWERING

Number of days taken for flowering in M₁ generation was 28-38 in control plants. In treated plants, it was 26-36 days. In M₂ generation these were 25-35 days in control plants. There were no significance change observed among the plants treated with different concentration of lead in control but initiation was first noticed in 01% in both generation M₁ and M₂. Thus it is clear

that single dose treatment of seeds with lead nitrate up to 01% concentration affect the flowering period in the case of *Phaseolus aconitifolium* among the survived plants in both generations i.e. M₁ & M₂ generations.

EFFECT ON NUMBER OF PODS PER PLANT

The average number of pods in *Phaseolus aconitifolium* is also declined with increase in the treatment concentration of lead nitrate. The average number of pods decreased to 24.0, 23.5, 21.0 & 19.5 in 0.1%, 0.25%, 0.50% & 01% treatment concentration of lead treated plants respectively in comparison to control which is 35.5 in M₁ generation. In M₂ generation, the average number of pods are 52.6, 48.6, 44.3 & 42.9 in 0.1%, 0.25%, 0.50% & 01% treatment concentration of lead nitrate while it is 51.8 in control.

Above observations shows that there is a declining trends in M₁ generation in each treatment concentration. However, in M₂ generation, the average number of pods in 0.1%, treatment concentration treated plants show slightly increase (1.4%) in the average number of pods but in other treatment concentration, the trend is declining.

When we consider the percentage decline, it is higher in M₁ generation and comparatively lower in M₂ generation. Moth bean developed adaptability in M₂ generation in comparison to M₁ generation.

The result is also supported by Nasralla and Ali, (1985) who have reported that deposition of Pb on vegetation growing along the road sides not only affects growth and germination but also fruit production of plants. Foliar applications of lead nitrate solution caused reduction in various growth indices and yield parameters of wheat (Rashid and Mukherjee 1990).

Soil contaminated with Pb cause sharp decrease in crop productivity thereby posing a serious problem for agriculture (Johnson and Eaton. 1980).

EFFECT ON DAYS TAKEN FOR HARVESTING

The harvesting period in M₁ generation was 62-65 days while it was 60-65 days in M₂ generation for both treated and untreated plants. Thus there is no significant change in the harvesting period in both generations in comparison to control.

EFFECT ON WEIGHT OF SEEDS

The average weight of 100 seeds were 1.420gm, 1.363gm, 1.346gm and 1.304gm in 0.1%, 0.25%, 0.50% & 01% treatment concentration of lead nitrate in M₁ generation in respect to 1.859gm in control. Reduction in weight was also noticed in M₂ generation. There were 2.241gm, 2.046gm, 2.037gm and 1.926 gm in 0.1%, 0.25%, 0.50% & 01% treatment concentrations of lead nitrate. Weight of control seeds was found to be 2.197 gm in M₂ generation.

However, percentage reduction in the weight of seeds obtained in M₁ generation is higher in the comparison to M₂ generation. The percent reduction in weight are 23.6%, 26.68%, 27.59% & 29.85% in 0.1%, 0.25% , 0.50% & 01% concentration lead treated plants while it were 6.87%, 7.28% & 12.33% reduction in the weight in 0.25%, 0.50% & 01% concentration lead nitrate treated plants in M₂ generation. There is a slight gain in weight of 3.68% in 0.1% lead treated plants.

Thus there is a loss in the yield under the stress of lead nitrate. The trend of the loss in yield is increasing with increase in the treatment concentration in both generations. But in M₂ generation the loss percentage is less in comparison to the M₁ generation. It is due to development of adaptability or stress tolerance capacity in successive generation.

Moth bean plants showed a decreasing trend with increasing concentration of lead nitrate treatment. It may be due to occurrence in changes in normal physiological functions and biochemical synthesis under stress condition of metal toxicity as was similarly reported by some workers (Kabir *et al.*, 2008; Buts *et al* 2013).

We have tried to assess the mutagenic effect of a single dose treatment given to seeds before sowing into fields on the growth & productivity of the *Phaseolus aconitifolium* up to two generations. Observations clearly indicate the toxicity of the lead nitrate. Single dose treatment

before sowing of seeds into field has reduced the germination & seedling survival. Further among survived plants, it adversely affects the morphological & reproductive features of the plants. Even it deteriorated the seed quality which was the final product of the crops. This happening clearly indicates the bio-accumulation & mutagenic effect of lead nitrate. Mutagenic effects and bio-accumulation of lead nitrate clearly reflects the health hazards of human beings as it will reach to the human body through food and may disturb the physiology & metabolism of human leading to the health hazards.

Therefore, there is a need of urgent attention to develop the system for monitoring & regulation of industrial & domestic effluents & providing appropriate advice and support as per requirements.

REFERENCES

1. Baki A.A. and Anderson J.D. (1973): Vigour determination in soyabean by multiple criteria. *Crop Sci.*, 13: 630-633.
2. Buts A.K., Chaudhary V.L. and Singh D. (2013): Morphology & productivity response of *Lens esculentus* (Lentil) under the stress of Cadmium chloride (CdCl₂). *India. J.L. Sci.*, 2(2): 23-26.
3. Dalal T. and Bairgi (1985): Effects of Hg, As and Pb on germination and seedling growth of two jute varieties. *Environmental Quality*, 2: 93-96.
4. Eun S., Youn H.S. and Lee Y. (2000): Lead disturbs microtubule organization in the root meristem of *Zea mays*. *Plant, Physiol.*, 110(3): 357-365.
5. Fargasova A. (1994): Effects of Pb, Cd, Hg, As and Cr on germination and root growth of *Sinapis alba* seeds. *Bull. Environm. Contam. Toxicol.* 52: 452-456.
6. Haghiri F. (1973): Cadmium uptake by plants. *J. Environ. Qual.*, 2: 93-96.
7. Johnson, M.S. and Eaton, J.W. (1984): Environmental contamination through residual trace metal dispersal from derelict lead-zinc mines. *J. Environ. Qual.* 9: 175-179.
8. Kabir M., Zafar Iqbal M., Shafique M. and Farooki Z.R. (2008): Reproduction in germination and seedling growth of *Thespesia populnea* L. caused by lead and cadmium treatments. *Pak. J. Bot.*, 40(6): 2419-2426.
9. Kabir M., Zafar M. and Shafique M. (2009): Effects of Lead on Seedling Growth of *Thespesia populnea* L. *Advances in Environmental Biology*, 3(2): 184-190.
10. Khajeh-Hussein M., Powell A.A. and Bigham I.J. (2003): The interaction between salinity stress and seed vigour during germination of soyabean seeds. *Seed Sci. Technol.*, 31: 715-725.
11. Kiran Y. and Munzuroglu O. (2004): The effects of lead on the seed germination and seedling growth of *Lens culinaris* Medic. *Firat University, J. Sci. Engineering*, 16(1): 1-9.
12. Mhatre G.N. and Chaphekar S.B. (1982): Effects of heavy metals on seed germination and early growth. *Environ. Biol.*, 3: 53-63.
13. Monzuuroglu O. and Geckil (2002): Effects of metals on seed germination, root elongation and coleoptiles and hypocotyle growth in *Triticum aestivum* and *Cucumis sativus*. *Arch. Environ. Contam. Toxicol.*, 43: 203-214.
14. Nasralla M.M and Ali E.A. (1985): Lead accumulation in edible proteins of crop grown near Egyptian traffic roads. *Agriculture Eco. Environ.*, 13: 73-82.
15. Nriagu J.O. (1992): Toxic metal pollution in Africa. *Sci. Total Environ.*, 121: 1-137.
16. Patra M., Bhoomik N., Bandhopadhyay B. and Sharma A. (2004): Comparison of mercury, lead and arsenic with respect to genotoxic effects on plant systems and the development of genetic tolerance. *Environ., Exp. Bot.*, 52: 199-135.
17. Pavivoke A.E.A. (2002): Soil lead alters phytase activity and mineral nutrient balance of *Pisum sativum*. *Environ. Exp. Bot.*, 48: 61-71.
18. Rashid P. and Mukherjee A. (1990): Effects of lead nitrate on some anatomical features of mung bean. *Bangladesh Journal of Botany*, 19: 146-154.
19. Shafiq M. and Iqbal M.Z. (2005): The toxicity effects of heavy metal on seed germination and seedling growth of *Cassia siamea*. *Lamk. J. News Seeds*, 7: 95-105.
20. Singh R.S., Rao A.S., Joshi N.L. and Ramakrishna Y.S. (2000): Evapotranspiration rates and water utilization of moth bean fewer than two soil moisture conditions. *Annals of Arid Zone*, 39(1): 1-8.
21. Wierzbica M. and Obidzinska J. (1998): The effect of lead on seed imbibitions and germination in different plant species. *Plant Sci.*, 137: 155-171.