

Heavy Metal Tolerance and Growth Responses of *Tagetes Patula* Grown in Fly Ash Amended Soil

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Abstract- – Coal based power generation is a principal source of electricity in India. About 15-30% of total amount of residue generated during coal combustion is flyash which contains many toxic heavy metals like Cr, Pb, Hg, and Cd along with essential elements like S, B, Ca, Na, Fe, Zn, Mn and P. Due to presence of hazardous metals fly ash disposal is a major environmental issue. To overcome this problem in present research phytoremediation based strategy is used in which we studied the effect of various concentration of fly ash (0%, 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80% 90% & 100%) on heavy metal tolerance and growth responses of *Tagetes patula* (French mix orange variety). Application of fly ash to soil has become an alternative to chemical fertilizers upto some extent due the presence of many essential elements(S,B,Na,Ca, P, Zn & Mn) into it . Results revealed that plants grown on soil amended with 10% fly ash have shown the maximum growth performance. However significant increase in plant biomass & plant height (shoot length, root length) is observed upto 30% fly ash amendment in comparison to control soil. Fly ash amended plants further amended with farmyard manure which slightly improve the performance of this plant. The results indicated that *Tagetes patula*(French mixed orange variety) may be used for phytoremediation of flyash contaminated soil.

Keywords:- Biomass, fly ash, coal, phytoremediation, heavy metals, *Tagetes patula*

I. INTRODUCTION

In India 75% of electricity is generated by coal based thermal power plants, which produce nearly 65 million tons of fly ash in a year as a by-product [1] and this flyash production will exceed 140 million tons per annum by 2020 [2, 3]. Out of this large scale production only a minor percentile of fly ash (<15%) is being used for preparing bricks, ceramics and cements [4] and major portion remains unused which is usually disposed in basins or landfills near the power plants. The fly ash disposal to the land poses environmental health hazards due to high concentration of its toxic heavy metals such as Cu, Cd, Ni, Pb, Cr etc. [5, 6]. A number of conventional methods have been developed to remediate metal contaminated soil, but these are encountered due to their adverse effect on soil structure & soil productivity, requirement of high energy input and expensive machinery [7]. In the recent years phytoremediation of fly ash contaminated soil is gaining a lot of importance since it is a cost effective, promising and environment friendly technology having extensive root system which extract and concentrate heavy metals from

the contaminated site and also provide a pleasant land scape. In phytoremediation, roots of the established plants absorb metal elements from the soil and translocate them to above ground shoot where they accumulate; the accumulated parts are harvested and removed resulting in permanent removal of metals from the site [8]. Translocation of metals was found to be more from roots to shoots in plants grown on fly ash amended soil [9]. Translocation of metal from roots to other parts of plants can be very useful in biological monitoring of heavy metal contamination as well as selection of metals accumulator or tolerant species [10]. Recently researchers are focusing the attention on selection of appropriate plants and appropriate amendments to the soil, to enhance the phytoremediation efficiency of the plants.

II. MATERIALS AND METHODS

A. Sample Collection and soil sample amendment with flyash

Fly ash samples were collected randomly from dumping sites of Jharli Thermal Power Plant, Jhajjar, Haryana, India, in large plastic bags and mixed together. The soil samples were collected from D.A.V. College of Engg. & Tech., Kanina (mohindergarh), Haryana. Fly ash as well as soil samples were air dried for 7 days and then passed through 2.37mm sieve. Then soil sample were amended with different proportions of flyash as detailed below: 10%(10% fly ash& 90% soil), 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90% , 100%. and soil served as control.

B. Reagents:

The analytical grade reagents were used. Certified aqueous standards of heavy metals were used to prepare the standards curve for AAS. The reagents, samples and standards were stored in polyethylene containers and preserved at low temperatures.

C. Physico-chemical Analysis of different amendments:

pH and EC of the samples were analysed by digital pH and conductivity meter (Model SLT-720 & SLT-615) by dissolving 10 gm of soil or fly ash in 100 ml of distilled water after proper stirring and settling. Organic carbon and organic matter were estimated using methods of Kalra and Maynard, 1991 [11].

For heavy metals analysis samples were digested on hot plate till brown fumes were stopped by using acid digestion technique in which 1 gm of fly ash sample was dissolved in 10 ml 3:1 percent solution of conc. HNO₃ and H₂SO₄. Samples then cooled and total 50 ml volume of each sample was made by using double distilled water. Filtered and stored in well washed plastic bottles and were analysed through AAS by standards methods.

D. Experimental design:

Pots were filled with 10% (10% fly ash & 90 % soil), 20%, 30%, 40%, 50%, 60%, 70%, 80% , 90% and 100% fly ash contaminated soil in triplicates along with one triplicate set of control soil.

The seeds of *Tagetes patula* (French mix orange variety) were sterilised with 0.1% mercuric chloride for 5 minutes to avoid fungal contamination, washed with distilled water and soaked in water overnight. Soaked seeds were evenly sown (@ 4 seeds/ pot) in all the pots in triplicate having control and fly ash contaminated soil. After one week seedling plants were thinned, so, that one plant remains in one pot and are kept under natural conditions for full developments, which are irrigated at regular intervals as and when required.

Similar experimental design was set with 10 gm of farmyard manure amendment in each control and fly ash contaminated pot.

E. Harvesting of the plants:

The fully grown plants of *Tagetes patula* (French mixed orange variety) were harvested from control soil; fly ash contaminated soil and farmyard manure amendment soil after 90 days and repeatedly washed with double distilled water.

F. Determination of metal tolerance:

Root length, shoot length, plant height, wet and dry biomass of harvested plants was measured for the

determination of metal tolerance in different fly ash contaminated samples and farmyard amended samples.

G. Digestion of harvested plant materials & soil samples:

Harvested plants were air dried for 3 to 5 days and then oven dried at 80 °C till constant weight attained. The root & shoot parts were separated and homogenized by grinding in a stainless steel blinder. Then grinded material was sieved from 2.37 mm sieve and kept for further analysis. One gram each of soil, roots and shoots were taken into a 100 ml beaker and digested into 42 ml of tri acid mixture (HNO₃: HClO₄: H₂SO₄ in 5:1:1 ratios) at 80 °C till the transparent solution appeared. The digested material was cooled, filtered with Whatmann no. 42 and maintained to 50 ml with distilled water in well washed plastic bottles and then were analysed for different heavy metals by AAS.

III. RESULTS AND DISCUSSION

Physico-chemical properties of different amendments of fly ash and control soil were presented in table-I. Data analysis revealed that the control soil used in this experiment was slightly acidic, while fly ash amended soils were alkaline in nature. The alkaline nature of fly ash amended soils may be due to the presence of CaO and MgO in fly ash. EC decreased with increase in flyash amendment ratio due to the precipitation of soluble cations by alkalinity of flyash in fly ash amended soil. Organic matter and organic carbon were found decreased with increase in flyash ratio.

The level of different heavy metals in fly ash and control soil was presented in table-II. The analysis of result showed the significantly higher level of all the heavy metals (Cr, Cd, Co, Mn, Ni, Cu, Pb) in fly ash than the control soil.

TABLE I. PHYSICO-CHEMICAL ANALYSIS OF CONTROL SOIL, FLYASH AND FLY ASH CONTAMINATED SOIL

S. No.	Physical Parameter	Soil	Flyash	Flyash contaminated soil (%)								
				10	20	30	40	50	60	70	80	90
1	pH	6.0	8.5	7.1	7.3	7.5	7.5	7.6	7.9	8.0	8.1	8.4
2	EC	750	320	690	650	620	580	530	480	400	340	320
3	Organic matter (%)	3.8	3.0	2.8	2.8	2.9	2.7	2.7	2.7	2.7	2.8	2.9
4	Organic carbon (%)	2.3	1.8	1.8	1.8	1.7	1.5	1.6	1.6	1.6	1.7	1.7

TABLE II. LEVEL OF HEAVY METALS CONCENTRATION (MG KG⁻¹ DW) IN FLYASH AND SOIL

S. No	Parameters	Flyash	Soil
1	Cr	21.7	14.3
2	Cd	1.87	1.35
3	Co	3.86	2.08
4	Mn	104.96	3.07
5	Ni	15	3.49
6	Cu	10.817	3.0
7	Pb	1.67	1.05

The fly ash amended soil showed the significant increase in the growth parameters such shoot length, plant height and plant biomass (wet) upto 30% fly ash amendment in comparison to control soil. This significant increase in all growth parameters is due to the presence of almost all the essential plant nutrients in fly ash which improve the soil texture and other soil property at lower fly ash amendments. After 30% fly ash amendments all fly ash amended soil showed significant reduction in all the growth parameters in comparison to control soil (Table-III).

The reduction in all the growth parameters clearly indicated that fly ash amendment has caused heavy metal stress in the soil environment. The fly ash contaminated soil was amended with 10 gm of farm yard manure and result analysis revealed a slight increase in all growth parameters in all fly ash contaminated soils (Table-IV).

TABLE III. EFFECTS OF FLYASH AMENDMENTS ON GROWTH PARAMETERS OF TAGETES PATULA (FRENCH MIXED ORANGE VARIETY)

S. NO.	Plant growth parameters	Control soil	Flyash	Flyash amendments (%)								
				10	20	30	40	50	60	70	80	90
1	Root length(cm)	12	6.5	13	12.6	12.5	11	9	7	6.8	6.8	6.7
2	Shoot length(cm)	50	20	54	53.8	53.6	40	35	26	21.5	21	20.5
3	Plant height(cm)	62	27	67	65	64	51	45	35	31	30	27
4	Wet biomass(gm)	52	24.5	60	56	52	40	37	34	32	30	26
5	Dry biomass(gm)	25	10.8	27	25	24	19	15	13.8	13.2	13	12.5

TABLE IV. EFFECTS OF FARMYARD MANURE AMENDMENTS ON GROWTH PARAMETERS OF TAGETES PATULA (FRENCH MIXED ORANGE VARIETY): EACH POT IS AMENDED WITH 10 GM OF FARM YARD MANURE

S. NO.	Plant growth parameters	Control soil	Flyash	Flyash amendments (%)								
				10	20	30	40	50	60	70	80	90
1	Root length(cm)	12.5	6.8	13	12.8	12.5	11.5	10	7	6.8	6.8	6.8
2	Shoot length(cm)	54	20.5	58	55	54	44	37	28	23	21.9	20.9
3	Plant height(cm)	65	27.4	70	67	66	55	48	35	31	30	27.8
4	Wet biomass(gm)	60	26.0	65	60	55	45	40	36	33	30.8	26.7
5	Dry biomass(gm)	28	11.2	29	26.5	25	21	17	14	13.6	13.4	13

IV. CONCLUSIONS

The present study revealed that the fly ash could be beneficial in improving the soil quality and plant growth. The most suitable treatment for improving the growth of French mixed orange variety of the Tagetes patula plant is 10%, fly ash with soil as it gives the maximum growth but the growth was found with significant improvement upto 30% flyash amendment with soil. This plant survive in all the fly ash amendments, which indicates that the French mixed orange variety of the Tagetes patula can tolerate heavy metal pollution and can be used as an ideal plant for phytoremediation of fly ash contaminated soil.

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