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Comparative productivity and Mycorrhizal infectivity of Fly Ash with other soils and standardization of amendment ratio of Fly Ash with lateritic soil

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Fly ash is a by-product of thermal power and its disposal has been highly problematic to our environment. Many researchers proposed to use fly ash with soil that may improve physical, chemical, biological properties and act as a source of readily available micro and macro nutrients to the plants. The samples were collected from new vegetation, old vegetation, and without vegetation in fly ash deposited area of Kolaghat thermal power station. Different soil samples from agricultural and forest lateritic soil and fly ash were compared for AM spore count, root colonization and productivity of sesame. The soil from potato field showed maximum productivity followed by fly ash from old vegetation. The treatment of fly ash from old vegetation, showed maximum spore number and root colonisation. At the second step a combined mixture of the lateritic soil and fly ash in different ratio were taken to standardize for amendment of lateritic soil. As test crop sorghum were grown. Plant fresh weight, root colonization percentage, spore number was measured. Among all different ratio of lateritic soil and fly ash 1:5 (v/v) showed the maximum fresh weight and AM-root infection intensity compared to other ratio and control. Hence reclamation of lateritic soil by fly ash may induce batter productivity.

Key words: Bioremediation, immobilization, mycorrhization, reclamation

INTRODUCTION

Fly ash is a particulate residue of coal based thermal power plants. In India total of power generations, 75% is produced by coal based thermal power plants and a huge amount of fly ash is being generated as byproduct from its depositions are becomes a real problem to environment at present. It causes air and water pollution if proper management measures are not taken in time.It is highly alkaline and rich in salts (Adriano *et al.,* 1978) and an amorphous mixture in large amount ferro-aluminosilicate of elements like C, K, Ca, Mg, Cu, and Zn (Raularay *et al.,* 2003; Lee *et al.,* 2006; Tiwari *et al.,* 2008). Fly ash also contains trace amounts of toxic heavy metals U, Th, Cr, Pb, Hg, Cd etc. which affects human health, plants and the environment. But fly ash has been noted for its potential use as a soil amendment (Wong, 1995) and can improve physical, chemical and biological properties of soils and is a source of readily available plant micro and macro nutrients. It contains many

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essential plant nutrients and could be a potential source of essential nutrients for plants (Pandey *et al.* 1994; Singh *et al.* 1997; Kuchanwar and Matte, 1997). The high concentration of elements like K, Na, Zn, Ca, Mg and Fe in fly ash increases the yield of many agricultural crops. Application of fly ash provided its positive value for crop growth.

Arbuscularmycorrhizal fungi (AMF) are widely occurring soil microorganisms that are obligately aerobic (Harley and Smith, 1983), forms symbiotic associations with plant roots have been shown to stimulate re-vegetation by supplementing the nutrient absorption capacity of the plant root systems, resulting in increased seedling survival and growth of the host plant (Perry and Amaranthus, 1990). They have been known to enhance crop growth and yield (Douds *et. al.*, 2005) through increased water and nutrient uptake, as well as alteration of some physiological processes in the plants that result in increased yield (Oyetunji *et. al.*,2003). Arbuscular mycorrhizal fungi accumulate heavy metals from fly ash. AM fungi have been used as bioremediation agents (Leyval *et al.*, 1997) and acts as biofertilizers for agricultural, horticultural and silvicultural plant species in polluted area (Lakshman, 2009). AM fungi helps in binding the fine particles of ash and arrests the uptake of heavy metals by host plants. AM fungi improved soil properties in stressed environments (Sarangi and Mishra, 1998; Ortega-Larrocea *et al.*, 2010). AM fungi are important components in re-vegetation of disturbed and potentially toxic environments because they can contribute to nutrient availability, immobilize heavy metals in the soil, and bind soil particles into stable aggregates that improve soil structure and reduce erosion potential. Some workers suggested that addition of fly ash up to 10% decreases the bulk density and increases the water holding capacity(Black, 1965). Alleviation of heavy metal phytotoxicity by AM fungi has been indicated in several studies (Chen *et al.*, 2007; Arriagada *et al.*, 2004). The AM fungi may enhance plant P nutrition and increase the plant growth by diluting metal effect in host plant or by binding of the metal to the fungal mycelium and immobilize them in rhizosphere or roots (Chen *et al.*, 2001).

Some different soil samples from forest and agricultural land are compared for AM spore count and root colonization and productivity of sesame grown in those soil.Also a greenhouse experiment has been conducted with sorghum plant to study the infective potential of rhizospheric AM fungus of the fly ash used as inoculum.

MATERIALS AND METHODS

Rhizospheric soil samples (without vegetation, new vegetation, and old vegetation) were collected from fly ash deposited area of Kolaghat thermal power station, Kolaghat, Purba Medinipur, West Bengal (22.41^0 N Latitude and 87.87^0 E Longitude). Soil samples were also collected from different areas-forest (lateritic zone) and agricultural land (paddy field and potato field)where no deposition was done.For analysis of plant growth, sesame plants (*Sesamum indicum*) under various soil conditions (T_1 : Fly ash without vegetation, T_2 : Fly ash with new vegetation, T_3 : Fly ash with old vegetation, T_4 : Soils from potato field, T_5 : Soils from paddy field, and T_6 : Soils from lateritic forest zone)were grown for 60 days.For study of this comparative productivity in fly ash and other soils, sesame was grown in polythene bags (20X20 cm.) with5 replicates.Growth performances of cesame plants under various soil conditions was studied in term of height, leaf number and leaf area. Data were recorded from 15th days of plantation (/sowing) of each treatments and continued up to 60th days after sowing (d.a.s.) with 15 days of interval. VAM spore numbers before and after sesame plantation and plant total dry weight were measured. Total number of spores was counted in 100 g soil.

For infection potential study and standardization of fly ash volume in soil, 100% control of collected lateritic soil and fly ash, and a combined mixture of the sterile lateritic soil and collected fly ash [F] (1:1, 1:2, 1:3, 1:4, and 1:5 v/v ratio of total 250 ml) were taken as treatment.*Sorghum* were grown in plastic pots of (18 x 7.5 cm) and 250 ml of volume were used and the design was with 3 replicates. After 60 days total plant fresh weight, root colonization percentage, spore number was measured. Fine tertiary root samples were carefully collected, treated with 10% KOH solution and stained with 0.5% cotton blue solution overnight. Fifty root pieces were examined for each sample and root colonization percentage was calculated. The root colonization percentage was calculated by the formula:

$$\text{Root colonisation percentage} = \frac{\text{Total No. of root pieces observed}}{\text{No. of root pieces colonised}} \times 100$$

Statistical analysis of data was done in term of correlation coefficient, least significant difference using IBM SPSS (v 19.1) software.

RESULTS AND DISCUSSION

In the first step of the experiment sesame plants grown in various soils. At 15 days after sowing, plant height was found maximum at T_5 (paddy field soil) followed by T_4(potato field soil) and T_3(fly ash from old vegetation) (Table 1). Total leaf number was maximum also at T_5 followed by T_4 and T_3 but maximum leaf areawas in T_4 followed by T_3 and T_2(fly ash with new vegetation), while minimum height was found in T_1 followed by T_2 On 30th day, the least height in T_1 increased 122.44%. The plant growth in term of height, leaf number and leaf area was became maximum in T_4 and minimum in T_6. In 45th day, that trend was followed and continued till 60 days after sowing. In 60thday, all growth parameters of T_3 was higher than others (T_1 T_2 T_5 and T_6), except only from T_4. T_6 showed least growth in 60th day, though at 15th day that was almost high, but rate of increment was least after then (in height and leaf number). Whereas T_1showed least growth in 15th day, but the rate of increment was maximum in height (324.4%) and leaf number(208.6%). In 60th day,

Table 1 : Growth parameters of sesame plants under various soil conditions

	15 d.a.s.			30 d.a.s.			45 d.a.s.			60 d.a.s.		
Treatments	Height (cm.)	Leaf Number	Leaf area (cm^2)	Height (cm.)	Leaf Number	Leaf area (cm^2)	Height (cm.)	Leaf Number	Leaf area (cm^2)	Height (cm.)	Leaf Number	Leaf area (cm^2)
										$20.8^{h}_{e^*}$	$14.2^{b}_{b^*}$	$5.8^{a\,a^*}$
T_1	4.9	4.6	4	10.9	7.2	4.8	15.3	10.2	5.4			
										[324.4 %]#	[208.6 %]#	[45%]#
										$21.7^{g}_{d^*}$	$14.6^{b}_{b^*}$	$6^{a\,a^*}$
T_2	5.2	5.2	4.4	10.5	7.8	5	15.8	10.4	5.2			
										[317.3 %]#	[180.7 %]#	[36.3 %]#
										$24.3^{c\,b^*}$	$17.8^{a}_{a^*}$	$6.2^{a\,a^*}$
T_3	5.9	5.8	4.6	11.1	7.6	5.2	16.2	11.6	5.6			
										[311.8 %]#	[206.8 %]#	[34.7 %]#
										$26.7^{a}_{a^*}$	$19^{a\,a^*}$	$6.8^{a\,a^*}$
T_4	6.3	6.2	5.4	12.8	8	6	20.1	13	6.4			
										[323.2 %]#	[206.4 %]#	[25.9 %]#
										$23.5^{d\,c^*}$	$17^{a\,a^*}$	$5^{a\,a^*}$
T_5	6.9	6.6	4.2	11.6	7	4.4	16.8	11.8	4.8			
										[240.5 %]#	[157.5 %]#	[19%]#
										$17.9^{k\,g^*}$	$11.8^{c\,b^*}$	$3.4^{b\,b^*}$
T_6	5.8	5.6	2.4	9.1	5.8	2.4	14.3	8.2	3			
										[208.6 %]#	[110.7 %]#	[41.6 %]#

Note: # Percentage (%) increased from 15 d.a.s. ; Data with same letters are significant at 5% level (a, d, c, d,g, h, k) and 1% level (a*, b*, c*, d*, e*, g*).

Table 2 : Spore numbers, root colonisation status and plant dry weight of Sesame plantunder various soil conditions

Treatments	VAM spore numbers (100 g. soil)		VAM-root colonization [60 d.a.s.]		Dry weight (g)
	Before plantation	After plantation	Infection %	Infection class	[60 d.a.s.]
T_1	17	28 [64.7 %]#	13	I	2.13 i f*
T_2	721	1108 [53.6 %] #	73	II	2.58 f d*
T_3	1270	1780 [40.1 %] #	88	II	3.13 c b*
T_4	222	563 [153.6 %] #	51	I	3.65 a a*
T_5	114	360 [215.7 %] #	48	I	2.18 i e*
T_6	338	479 [41.7 %] #	61	I	1.98 j f*

Note: # Percentage (%) increased. ; Data with same letters are significant at 5% level (a, c, d,e, f, i, j) and 1% level (a*, b*, d*, e*, f*).

the height of T_4 wassignificantly higher ($P<0.05$ and $P<0.01$) than other treatments. In case of leaf number there were no significant difference between T_3 T_4 and T_5 and betweenT_1 and T_2 ($P<0.05$). At 60th day leaf area was maximum in T_4 followed byT_3 T_2 T_1 and T_5 but there was no significant difference 'among them ($P<0.05$ and $P<0.01$).

On 60 days after sowing, the dry weight of T_4 wassignificantly higher ($P<0.05$ and $P<0.01$) than other treatments (Table 2). The spore population was maximum in T_3 followed by T_2 before and after plantation This result may be due to hugevolume for light weight of fly ash. The percentage of increase in spore number was highest in T_5 (215.78%) followed by T_4 (153.6%) and minimum inT_6 (40.71%) Root colonisation percentage was found maximum in T_3 followed by T_2 and T_6. Less root colonisation percentage in agricultural soil may be due to agrochemicals used (Ghosh, 2007; Jasper *et al.*, 1979; De Miranda and Harris, 1994; Shen *et al.*, 1994). Fly ash from old vegetation (T_3) showed maximum spore number and root colonisation, where its induce productivity wasonlyless than potato field soil (T_4). Spore population of all treatments (T_1 T_2 T_3 T_4 T_5 andT_6) were found positively correlated 'with their dry weight ($r=0.49$). But root colonisation showed no any positive significant correlation with dry weight. Soil from potato field contained heavy nutrients used for previous crop. The residual effect is depicted in this result. But in soil from paddy field soil and lateritic soil, nutrient mobility is poorer than fly ash from old vegetation; that was reflected in plant

Table 3 : AM-root colonisation percentage of *Sorghum* under various soil conditions

Treatments (v/v ratio of Lateritic soil + Fly ash)	VAM-root colonization								Fresh weight (g.) [60 d.a.s.]	VAM spore numbers (100 g. soil)	
	15 d.a.s.		30 d.a.s.		45 d.a.s.		60 d.a.s.			Before plantation	After plantation
	Infection %	Infection class	Infection %	Infection class	Infection %	Infection class	Infection %	Infection class			
1:1 (L:F)	0	-	09	I	22	I	35	I	$4.2^{b\ a^*}$	2,571	2,702 [5.09%]#
1:2 (L:F)	04	I	17	I	31	I	48	I	$4.8^{b\ a^*}$	3,428	3,614 [5.42%]#
1:3 (L:F)	09	I	25	I	39	I	51	II	$5.6^{a\ a^*}$	3,625	3,857 [6.4%]#
1:4 (L:F)	14	I	29	I	45	II	60	II	$6.0^{a\ a^*}$	4,114	4,358 [6.58%]#
1:5 (L:F)	17	I	38	II	54	II	73	II	$6.1^{a\ a^*}$	4,285	4,637 [8.21%]#
100 % FLY ASH	21	I	49	II	69	II	85	II	$5.8^{a\ a^*}$	5,142	5,479 [6.55%]#
100 % LATERITE SOIL	15	I	23	I	47	I	61	I	$4.5^{b\ a^*}$	365	401 [9.86%]#

Note: # Percentage (%) of increase. ;
Data with same letters are significant at 5% level (a, b) and 1% level (a*).

growth parameters. Fly ash without vegetation showed least effect among three fly ash treatments, that indicates AM colonization and/ or prior phytoremediation has enhance the productivity. The dry weight in plants indicates the effective nutrient source from fly ash as all three treatments induced higher growth than paddy field soil and lateritic soil.

For standardization of the amendment ratio of fly ash with lateritic soil, among the different ratio used as treatments of L: F (v/v); 1:1, 1:2, 1:3, 1:4, and 1:5;it was found that 1:5 shows the maximum root colonisation percentage and fresh weight followed by 1:4, though no significant difference was present ($P<0.05$ and $P<0.01$) (Table 3). The spore number was found maximum in that ratio (except 100% fly ash control). Spore population of those treatments showed significant positive correlation with their fresh weight ($r=0.77$). Thus AM root colonisation shows the positive influence on the fly ash. From this experiment it was found that, the desirable ratio for laterite soil reclamation would be 1:5. The colonisation spore number and the fresh weight indicates this ratio highly acceptable and fittest.

The AMF helps in binding the fine particles of fly ash and arrest the movement heavy metalsand also helps in uptake of micronutrients and phosphorus solubilization (Adholeya, 2000). AM fungi improved the growth, physical properties of fly ash, and reduction of toxic metals (Juwarkar and Jambhulkar, 2008). AM fungi may suppress the uptake of Al, Fe, and Mn that may be present in toxic levels in some soils (Ning, 2000).Increased concentration of fly ash increases the plant growth as well as mycorrhizal status in root and rhizosphere of all the experimental plants. Increased tolerance of mycorrhizal plants to toxic heavy metal concentra

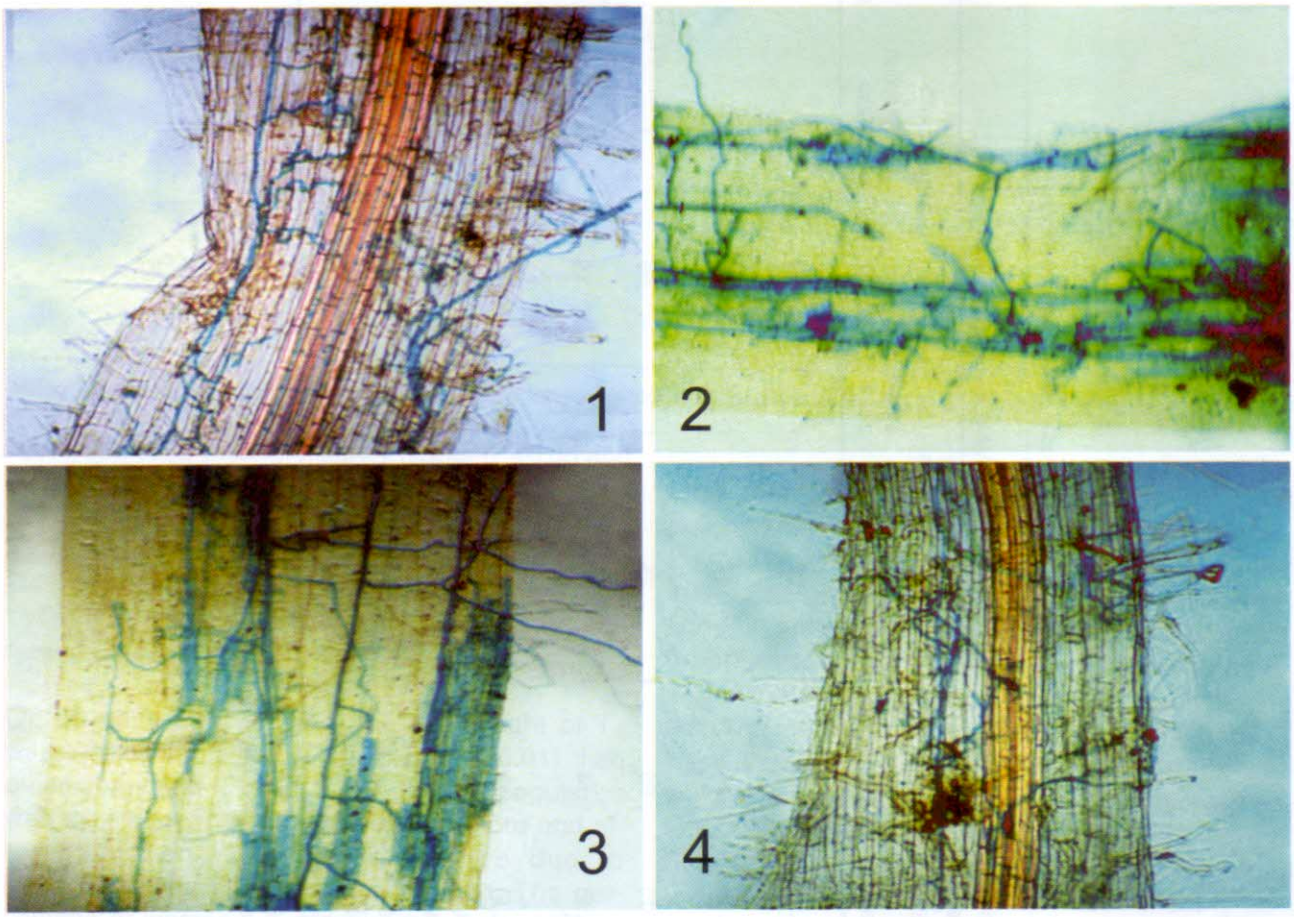

Fig. 1 : Root Colonisation: **1.**Root colonisation inthe treatment of 100% 1:4 (L: F) in 60 d.a.s., **2.**Root colonisation in the treatment of 1:5 (L: F) in 60 d.a.s., **3.**Root colonisation in the treatment of 100% fly ash in 60 d.a.s., **4.**Root colonisation in the treatment of 100% lateritic soil in 60 d.a.s.

tion in the soil makes mycorrhizae significant. Therefore fly ash may be used as a nutrient in agriculture or horticulture and also as a limiting agent in acidic agricultural soil (Plank *et al.*,1975; Sheela and Sundaram, 2003).

AM may have a major role in this process of increasing nutrient mobility and soil aggregation as in older vegetation shows high AM colonization and spore density.This study reveals that as a rich source of nutrient, fly ash specially, from older vegetation may be a good soil amending agent, particularly for nutrient poor lateritic soil.Fly ash specially, from older vegetation with high AM colonization is also a good source of bioremediation by mycorrhizae.

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