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Effect of fly ash application on soil microbial response and heavy metal accumulation in soil and rice plant



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ABSTRACT

Fly ash (FA), a byproduct of coal combustion in thermal power plants, has been considered as a problematic solid waste and its safe disposal is a cause of concern. Several studies proposed that FA can be used as a soil additive; however its effect on microbial response, soil enzymatic activities and heavy metal accumulation in soil and grain of rice (cv. Naveen) to fly ash (FA) application was studied in a pot experiment during dry season 2011 in an Inceptisol. Fly ash was applied at a rate of zero per cent (FS), five per cent (FA5), ten per cent (FA10), twenty per cent (FA20), 40 per cent (FA40) and 100 per cent (FA100) on soil volume basis with nitrogen (N), phosphorus (P) and potassium (K) (40:20:20 mg N:P: $K kg^{-1}$ soil) with six replications. Heavy metals contents in soil and plant parts were analysed after harvest of crop. On the other hand, microbial population and soil enzymatic activities were analysed at panicle initiation stage (PI, 65 days after transplanting) of rice. There was no significant change in the concentration of zinc (Zn), iron (Fe), copper (Cu), manganese (Mn), cadmium (Cd) and chromium (Cr) with application of fly ash up to FA10. However, at FA100 there was significant increase of all metals concentration in soil than other treatments. Microorganisms differed in their response to the rate of FA application. Population of both fungi and actinomycetes decreased with the application of fly ash, while aerobic heterotrophic bacterial population did not change significantly up to FA40. On the other hand, total microbial activity measured in terms of Fluorescein diacetate (FDA) assay, and denitrifiers showed an increased trend up to FA40. However, activities of both alkaline and acid phosphatase were decreased with the application of FA. Application of FA at lower levels (ten to twenty per cent on soil volume basis) in soil enhanced micronutrients content, microbial activities and crop yield.

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1. Introduction

Fly ash (FA), a byproduct of coal combustion in thermal power plants, has been considered as a problematic solid waste and its safe disposal is a cause of concern. FA being a coal combustion residue shows a wide variation in their physico-chemical and mineralogical properties depending on the nature of parent coal, conditions of combustion, type of emission control devices, storage and handling methods (Jala and Goyal, 2006). The major matrix elements in FA are Si, Al, and Fe together with significant percentages of Ca, K, Na and Ti. Ca was found to be the dominant cation in FA followed by Mg, Na and K (Matti et al., 1990). Al in FA is mostly bound in insoluble aluminosilicate structures, which considerably limits its biological toxicity. Fly ash contains

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http://dx.doi.org/10.1016/j.ecoenv.2014.03.033 0147-6513/© 2014 Elsevier Inc. All rights reserved. substantial quantities of trace metals (Cu, Zn, Mn, and Mo) and toxic elements such as vanadium (V), selenium (Se), arsenic (As), boron (B), aluminium (Al), Cd, lead (Pb), mercury (Hg) and Cr (Gupta et al., 2002), and exhibits metal toxicity in plants (Pandey et al., 2010). The pH of FA can vary from 4.5 to 12.0 depending largely on the sulphur content of the parent coal and the type of coal used for combustion affects the sulphur content of FA. The adverse environmental impact of dumping FA is contamination of soil and water with toxic and radioactive elements, land degradation and air pollution (Mallik, 2011).

The coal based thermal power plants, that constitute about 60– 70 per cent of total power generation capacity in India, use mostly bituminous coal with low calorific value and high ash content (35– 45 per cent), which lead to production of large volume of fly ash (112 mt) per year (Alam and Akhtar, 2011). In the current scenario of national dependence on coal for power generation, this figure may increase further in the years to come. Considering the high disposal cost (Rs. 50–100/mt) of FA and large area of land required for its disposal, it is imperative that the generated FA be utilized to the maximum extent (Dhadse et al., 2008). Considerable research has been focused on management and alternate use of fly ash in manufacture of cement, bricks, land filling, fertilizer fill, etc. Rice is an important crop of Indian agriculture grown on a variety of soil in different ecosystems. Due to ever increasing demand for food and shrinking of cultivable land resources, there is a need to produce more and more food per unit area which has made agriculture heavily dependent on chemical fertilizers. The indiscriminate use of chemical fertilizers affects soil health and, leads to a negative impact on soil productivity by eliminating diverse types of beneficial micro-organisms (Singh et al., 2010). In recent years, for sustainable productivity and improving soil health FA amendment is gaining importance in rice agriculture (Singh et al., 2011). Though, utilization of FA in agriculture is limited because of its low N and P contents, low soil microbial activity, high pH (Wong and Wong, 1989); there are some reports which mention potential use of FA as a soil ameliorant for improving physical properties of soil (Shen et al., 2008), as a liming material (Kumar and Singh, 2003) and a source of available plant micro- and macro-nutrients (Rautaray et al., 2003). Besides this fly ash is also used for land filling in the low lying fields which are subsequently used for agriculture and forestry purposes, application at such higher rate modify the soil and fly ash ratio and may have consequences on enzymatic activity and nitrogen transformation (Pati and Sahu, 2004). Due to the low bulk density and nutrient contents, generally the FA is applied in huge quantities to the agricultural fields. It has been reported that the application of FA from 10 to 200 t ha⁻¹ did not show toxicity effects to the rice plants and increased the yields of paddy (Bhaskarachary et al., 2012; Lee et al., 2006). Most of the studies on fly ash are mostly focused on its impact on plant growth and productivity, heavy metal accumulation in plant and management practices to minimize the adverse impacts of FA. However, the impact of FA on soil fertility, soil microbial/biochemical activity and soil nitrogen cycling is very limited (Pandey and Singh, 2010). Therefore, there is a need to thoroughly test the extent of heavy metal accumulation in soil and its overall impact on soil biological health, rice plant growth and heavy metal contents in order to establish an eco-friendly FA dose for safe soil amendment. Hence, the objectives of the present study are i) to investigate the impact of fly ash amendment on microbial responses in soil by measuring selected microbial populations, processes, and enzyme activities., and ii) to assess the extent of heavy metal accumulation in soil and rice plant due to application of FA.

2. Methodology

2.1. Experimental setup

A pot culture experiment was conducted in the dry season of 2011 with rice (cv. Naveen) in the net house of Central Rice Research Institute, Cuttack, India. The soil used in the experiment was an Aeric Endoaquept. The fly ash was collected from the FA-dykes of Aarti Steel Plant, Athagarh, Odisha, India and this fly ash was stabilized at 52 °C for 24 h to kill off any pathogens followed by drying at room temperature for one week. After the stabilization period, the mixtures were ground to pass through a 4 mm sieve to obtain homogeneous samples before mixing with the soil.

The fly ash and time-zero soil samples were analysed for pH (1:2), electrical conductivity (1:2), organic matter content, percentages of sand, silt, and clay, and extractable elements (Table S1, in the Supplementary materials section) using methods as described by Jackson (1973). The soil was thoroughly mixed, dried, ground and sieved through a 2.0 mm sieve and filled in earthen pots of 40 cm height and 30 cm upper diameter lined with polythene sheet along with FA and made up to 10 kg. The levels of FA were zero per cent, five per cent, ten per cent, twenty per cent, 40 per cent and 100 per cent on soil volume basis which were represented as treatments FS, FA5, FA10, FA20, FA40 and FA100, respectively. Each treatment was replicated six times of which three pots were used for microbial

analysis at panicle initiation stage (65 days after transplanting) and remaining three were taken up to harvest of the rice crop. Each pot was planted with 25 days old single rice seedlings and applied with recommended dose of NPK (40:20:20 mg N:P:K kg⁻¹ soil) to all the treatments. N was applied in two equal splits, one at basal and other at 60 days after transplanting. Destructive soil sampling from three randomly selected replicated pots of each treatment was done at panicle initiation stage for analysis of microbial parameters.

The soils collected from each pot were hand mixed, part of the soil sample was air dried and ground to pass through a 2 mm sieve and analysed for heavy metals. The remainder of the samples was placed in a plastic bag and stored in a refrigerator at 4 $^{\circ}$ C prior to microbial and biochemical analysis. All microbiological and biochemical analyses were performed within 5 days of sampling. Leaf area index was measured at flowering stage. Leaf area was recorded by putting each fresh leaves of one hill flat in a digital Leaf Area Meter (LI-3100, LiCor Inc., Lincoln, Nebraska). The area thus obtained was divided by the area of ground to get leaf area index. Observations on grain yield and other growth parameters were recorded at physiological maturity stage and plant samples were collected for analysis of heavy metals contents. Moisture percentage of the grains was determined using Infrared moisture content.

2.2. Estimation of microbial populations

Aerobic heterotrophic bacteria were enumerated by plating soil dilutions to agar media in petridishes. The agar medium used contained dilute (1:100 full strength) trypticase soy broth (Difco), ten per cent soil extract (prepared as described by Zuberer (1994)), and 1.5 per cent agar (difco). Plates were incubated at 28 ± 2 °C for 72 h prior to enumeration. Ammonium oxidizers and dentrifiers were enumerated using the MPN techniques of Schmidt and Belser (1994) and Tiedje (1994), respectively.

2.3. Measurement of soil enzyme activities

Knowing the sources of specific soil enzyme activities would greatly enhance our understanding of which group(s) of organisms are directly accessing a given nutrient resource particularly N and P, thus providing greater insight into the pathways by which energy and nutrients flow through the soil food web. Activities of alkaline and acid phosphatase were determined spectrophotometrically by measuring intensity of yellow colour due to formation of p-nitrophenol from pnitrophenyl phosphate (Tabatabai, 1994); urease enzyme activity was measured by Douglas and Bremner (1971) method which involves spectrophotometric estimation of unhydrolyzed urea after an incubation period. Fluorescein diacetate (FDA) hydrolysis assay was conducted spectrophotometrically by measuring formation of fluorescein (fluorescent yellow-green) to assess the overall enzyme activity of total microbial population (Adam and Duncan, 2001).

2.4. Nitrogen mineralization potential, nitrification potential and denitrification activity in soil

Nitrogen mineralization potential was determined by estimating the anaerobic production of ammonium (Bundy and Meisinger, 1994). Nitrification potential was determined using an aerobic incubation procedure (Schmidt and Belser 1982) in which a solution containing 50 mg L⁻¹ NH₄–N was added to 100 g freshly collected air dried soil contained in a glass beaker of 500 mL capacity and was kept in the incubator for a period of three weeks at 25 °C, another 100 g soil (without NH₄–N solution) was incubated separately as control. Moisture content of soil was maintained at field capacity (by periodic weighing and offsetting the loss of moisture) throughout the incubation period. The amount of NO₃–N content of soil at the beginning and end of incubation period was determined by extracting soil with 2 M KCl followed by a steam distillation method, and increase in NO₃–N concentration during the incubation period was expressed as nitrification potential of soil.

Denitrification activity was determined following the procedure described by Smith and Tiedje (1979); 20 g of freshly collected homogenized soil sample from each treatment was placed in the conical flasks of 250 mL capacity. 10 mL of deionized water and 100 mg of chloramphenicol was added to the soil and content was mixed to get slurry. The flasks were made air tight by sealing the mouth with a rubber septa through which the needle of syringe could pass, the headspace of the flask was evacuated and filled with O₂ free N₂ gas for three times to make the sample anaerobic and approximately ten per cent of the headspace was replaced with acetylene gas (C₂H₂). 10 mL of a solution containing of 56 mg KNO₃–N L⁻¹ was added and the flasks were shaken for 30 min to establish equilibrium between dissolved and gaseous N₂O. Gas samples were analysed in a gas chromatograph (Thermo CERES 800 plus GC) for determination of nitrous oxide using a Porapak Q column and an electron capture detector (ECD).

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