

STUDIES ON THE EFFECT OF ORGANIC AMENDMENTS AND EARTHWORM INOCULATION ON SOME IMPORTANT QUALITY PARAMETERS OF IRON MINE SPOIL

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KEY WORDS

Iron Mine Spoil
Organic Amendment
Bacterial Load
Soil Respiration
Earthworm

Received on :

05.10.2010

Accepted on :

23.12.2010

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ABSTRACT

Considerable quantity of mine spoil is generated during open cast mining of iron ore. Important physicochemical characters, bacterial load and heterotrophic respiration of iron mine spoil with and without organic amendment supplemented with earthworm (*Eisenia foetida*) inoculation have been studied over an incubation period of 28 days. Amended soil indicated enhanced pH, % organic C, P, N and K with respect to control. FYM treated spoil with worm indicated the highest (297 ± 14.3 cfu/g) bacterial count and heterotrophic soil respiration (29.8 ± 1.6 mg/m²/hr) followed by poultry manure amended spoil. The least values were obtained from unamended spoil. Significant ($p < 0.01$) difference between treatments and days of incubation for physicochemical parameters, bacterial load and soil respiration was observed. Earthworm inoculation enhanced bacterial load and respiration in both FYM and poultry manure amended iron mine spoil.

INTRODUCTION

Mining activities generally involve extraction or removal of minerals and metals from earth. Earlier, mining activities only considered the techno economic aspects. However, the environmental consequences to mining are now a matter of great concern. Large scale excavation is often necessary to extract a small amount of ore. Ore extraction disrupts the top soil besides causing serious damage to the flora and fauna (Cottard, 2001; Lin *et al.*, 2004).

The state of Orissa occupies an important position in the mining map of India with about 16.92% of the total mineral reserves of the country. The state has a resource of 53.05 million ton of iron ore (Directorate of Geology, Orissa, 2007-08). All iron ore mining operations in the state are open cast which produce large quantity of overburden (spoil) which are dumped in open. The spoil dumps are exposed to rains during monsoon resulting in leaching of the major soil nutrients. Furthermore, the iron mine spoil generally indicate significantly low microbial load and microbial enzyme activities (Rath *et al.*, 2010).

The micro flora such as bacteria, fungi and actinomycetes, as well as soil invertebrates particularly earthworm, play significant role in decomposition and mineralization of dead organic matter in soil (Macfayden, 1970; Lee, 1985; Swift and Hopkin, 1996). Besides, the earthworms stimulate microbial action by increasing the surface area for microbial colonization of

substrate (Edwards and Fletcher, 1988; Winding *et al.*, 1997). Previous studies have shown that earthworm activity promotes organic matter decomposition significantly (Parmelee and Alston, 1986). Restoration of mine spoil needs organic input. High quality organic matter could supply substantial amount of nutrients (Lal, 1991; Palm *et al.*, 1997) coupled with improved biological activity (Winding *et al.*, 1997).

Many studies have shown the beneficial effect of earthworm on the soil through higher microbial population and soil metabolism (Mc Lean *et al.*, 2006; Herry *et al.*, 2008). The densities of pseudomonads and filamentous actinomycetes were consistently higher for rhizosphere soils augmented with earthworm *Lumbricus terrestris* (Elmer, 2009). It has also been reported that earthworms stimulate growth of nitrifying and methanotrophic bacteria in soil (Herry *et al.*, 2008) However, combined results of the different organic amendments with worms on mine spoil are limited. With the above backdrop, this study aimed at to study the effect of organic amendment and inoculation of an epigeic species of earthworm *Eisenia foetida* on some major physicochemical parameters such as % organic C, N, P and K, bacterial load and heterotrophic respiration of the iron mine spoil.

MATERIALS AND METHODS

Collection of samples

Table 1: Mean \pm SD values of some physicochemical parameters

Treatments Days	C			F1			F2			P1			P2							
	7	14	21	28	7	14	21	28	7	14	21	28	7	14	21	28				
pH	5.48 ± 0.01	5.17 ± 0.03	5.23 ± 0.34	5.25 ± 0.41	6.97 ± 0.11	7.11 ± 0.15	6.92 ± 0.23	6.88 ± 0.36	7.18 ± 0.01	6.54 ± 0.02	6.73 ± 0.11	6.62 ± 0.03	7.02 ± 0.12	7.07 ± 0.01	6.77 ± 0.13	6.58 ± 0.02	6.96 ± 0.14	6.97 ± 0.01	6.64 ± 0.04	6.57 ± 0.04
C(%)	0.24 ± 0.03	0.10 ± 0.01	0.20 ± 0.02	0.12 ± 0.03	0.51 ± 0.03	0.51 ± 0.02	0.76 ± 0.01	0.84 ± 0.01	0.48 ± 0.04	0.38 ± 0.05	0.50 ± 0.06	0.51 ± 0.03	0.38 ± 0.02	0.37 ± 0.02	0.342 ± 0.02	0.46 ± 0.01	0.32 ± 0.02	0.36 ± 0.03	0.52 ± 0.01	0.51 ± 0.02
P(kg/ha)	8.00 ± 0.05	8.00 ± 0.06	8.05 ± 0.07	7.12 ± 0.01	11.02 ± 0.01	13.02 ± 0.03	13.04 ± 0.03	15.02 ± 0.03	13.11 ± 0.21	11.02 ± 0.01	10.02 ± 0.03	9.08 ± 0.04	11.02 ± 0.01	11.05 ± 0.02	11.07 ± 0.02	11.08 ± 0.03	10.02 ± 0.01	9.08 ± 0.01	9.05 ± 0.02	9.07 ± 0.03
K(Kg/ha)	92.20 ± 0.15	87.03 ± 0.11	108.0 ± 0.34	85 ± 0.31	111 ± 0.78	113 ± 0.81	118 ± 0.87	121 ± 0.91	105 ± 1.11	103 ± 1.31	98 ± 1.21	94 ± 1.13	107 ± 1.21	105 ± 1.01	103 ± 1.03	98 ± 1.03	101 ± 1.11	101 ± 1.01	98 ± 1.21	95 ± 0.34

C = control; F1 = F1: mine spoil + FYM + earthworm; F2: mine spoil + poultry manure + earthworm; P1: mine spoil + poultry manure + earthworm; P2: mine spoil + poultry manure

The spoil samples were collected from the iron mining area of Joda Barbil block in Keonjhar district of Orissa. The samples were collected at random from a two years old spoil dump in the month of September, 2009. Farm yard manure (FYM) and poultry manure were obtained respectively from the dairy and poultry farms of College of Veterinary Science and Animal Husbandry, Orissa University of Agriculture and Technology (OUAT), Bhubaneswar. *Eisenia foetida* worms were collected from the vermiculture unit located in the OUAT farm.

Organic amendments

For organic amendment, 400g each of dried and powdered FYM and poultry manure was mixed thoroughly with 1.6 kg of sieved mine spoil. Each amendment was taken in triplicates in polythene bags. Three sets of control (without amendment) were also taken for comparative study. The control and amended samples were kept at room temperature for incubation purpose. The moisture of all the sets was maintained at 80% level by regular sprinkling of double distilled water. One set (in triplicate) in each of the amended spoil types was inoculated with 10 *Eisenia foetida* worms and the other set (in triplicate) of both the amended types was maintained without worm.

The treatments were as follows:

C1: control spoil (no amendment)

F1: mine spoil + FYM + earthworm

F2: mine spoil + FYM

P1: mine spoil + poultry manure + earthworm

P2: mine spoil + poultry manure

After incubation for 3 days the first set of spoil samples were collected from the polypots for physicochemical and microbiological studies. Thereafter the samples were collected at an interval of 7 days for a total period of 28 days for analysis.

Physicochemical studies

Physicochemical characteristics of all the samples were done for pH, % organic C, N, Phosphorus and Potassium. For the study of these parameters, samples were thoroughly air dried and crushed with wooden pestle and sieved through 2mm sieve. Soil pH was measured with the help of a digital pH meter. Percent organic C was measured by the method described by Walkey-Black (1934). Soil phosphorus and potassium and nitrogen were assessed by Olsen method (1954), Lu (1999) method and semi-micro Kjeldahl method prescribed by Chapman and Pratt, (1979) respectively.

Microbiological studies

Bacterial population of all the treatments was studied by dilution plate count method using Nutrient Agar and expressed as cfu/g soil (ICMSF, 1978).

Soil metabolism

Soil metabolism in terms of soil respiration was measured by Alkali trap method (Carter, 1993; Nay *et al.*, 1994).

Statistical analysis

One way ANOVA test was done to test the significance of difference ($p < 0.01$) for various parameters using M Stat C Software (Michigan State University, USA).

RESULTS AND DISCUSSION

The values of pH, % organic C, N, P and K have been presented in Table 1. It was observed that spoil amended with 20% FYM and poultry manure showed an increase in pH. The maximum pH of 7.58 was observed in FYM (without worms) i.e. F2 on the 7th day of incubation. The minimum pH value of 5.17 was observed in control spoil. FYM and poultry manures in general enhanced the pH value of amended spoils. The condition changed from an acidic range to near neutral indicating that 20% manure amendment creates a favourable condition for microbial growth. Percent organic C showed the maximum value in F1 on 21st day on incubation and lowest was observed on the same day of incubation in the unamended spoil. Percent organic C level increased from day 1 to 21st day of incubation in both the unamended and amended sets with worms and thereafter declined on the 28th day of incubation. Identical trend was also noticed for phosphorus and potassium values. The level of nitrogen was consistently higher in manure amended soil samples with respect to control. Table 2 depicts the bacterial population (1×10^2 cfu/g soil) in the control and amended spoil at different time intervals. The highest bacterial count of 297 ± 14.3 cfu/g was recorded on the 28th day of incubation in F1 and the lowest in C1 on the same day of observation. The bacterial population was significantly higher in all the amended sets with respect to control. It was further observed that, in both the amended sets with worms, the bacterial population count steadily increased from the day 1 to 28th day of incubation. In the amended sets without worms

Table 2: Mean \pm SD of bacterial population ($\times 10^2$ cfu/g soil)

Days of incubation					
Treatments	1	7	14	21	28
C1	80 \pm 2.71	63 \pm 1.8	62 \pm 1.6	46 \pm 1.2	43 \pm 1.4
F1	228 \pm 11.8	262 \pm 13.8	271 \pm 11.9	283 \pm 14.2	297 \pm 14.3
F2	121 \pm 10.2	128 \pm 9.6	115 \pm 12.3	108 \pm 10.2	105 \pm 9.8
P1	214 \pm 14.8	249 \pm 15.2	268 \pm 13.1	271 \pm 10.4	281 \pm 13.2
P2	135 \pm 9.8	121 \pm 8.8	113 \pm 7.6	109 \pm 8.9	103 \pm 7.8

C = control; F1 = F1: mine spoil + FYM + earthworm; F2: mine spoil + FYM; P1: mine spoil + poultry manure + earthworm; P2: mine spoil + poultry manure

Table 3: Mean \pm SD of microbial respiration (CO_2 evolution in $\text{mg/m}^2/\text{hr}$)

Days of incubation					
Treatments	1	7	14	21	28
C1	12.6 \pm 0.8	11.4 \pm 1.0	10.5 \pm 0.8	11.1 \pm 1.2	9.5 \pm 0.6
F1	23.4 \pm 1.5	25.2 \pm 1.6	28.2 \pm 0.9	29.7 \pm 1.7	29.8 \pm 1.6
F2	21.8 \pm 1.2	19.7 \pm 1.2	18.6 \pm 0.8	17.4 \pm 1.6	16.7 \pm 0.9
P1	21.5 \pm 1.2	26.7 \pm 1.4	26.8 \pm 1.3	27.2 \pm 1.8	27.8 \pm 1.5
P2	19.9 \pm 2.0	18.6 \pm 1.8	17.4 \pm 2.2	15.6 \pm 1.6	14.7 \pm 1.4

C = control; F1 = F1: mine spoil + FYM + earthworm; F2: mine spoil + FYM; P1: mine spoil + poultry manure + earthworm; P2: mine spoil + poultry manure

Table 4: Analysis of variance for control and amended spoil samples

Parameters	CD			F			CV (%)
	D	T	DT	D	T	DT	
pH	0.08*	0.08*	0.17*	47.84*	277.56*	13.5*	1.56*
% C	0.038*	0.04*	0.09*	130.57*	98.83*	31.48*	11.03*
P	7.97*	8.91*	17.82*	83.69*	108.28*	22.34*	14.97*
K	0.98*	1.09*	2.19*	463.14*	3547.85*	964*	0.32*
Bacteria	35.5*	35.5*	79.39*	1.09 ns	52.95	1.16 ns	28.31*
Respiration	1.53*	1.53*	3.42*	1.7 ns	144.54	2.14 ns	10.53*

D = days; T = treatments; * = significant at $p < 0.01$; ns = not significant

however, the bacterial count decreased after 7th day of incubation. In the control, the bacterial count declined from 80 \pm 2.71 cfu/g to 43 \pm 1.41 cfu/g during the experimental period of 28 days.

Long *et al.*, (2003) have reported that the total quantity of major soil microbes declined significantly in copper mine spoil compared to that of non mined soil. Similar results have been obtained by Pierzynski *et al.*, (1994); Machulla *et al.*, (2005) have reported that lower microbial biomass in mine spoil is largely due to low organic carbon content. In the present study, lower level of nutrients in unamended iron mine spoil might have been responsible for lower number of bacterial colonies in the control sets. Addition of organic manures, rich in nutrients possibly accelerated the growth of bacterial population in the amended sets.

The amended sets with worms exhibited higher bacterial count in comparison to the sets without worms. It is an established fact that epigeic worms such as *Eisenia foetida* play important role in the decomposition of organic matter and nutrient turnover in the soil (Lee, 1985; Winding *et al.*, 1997). Earthworm inoculation enhances bacterial count due to the fact that worm activity accelerates soil aeration process, thus increasing the number of aerobic microbes, which resulted in higher bacterial count in worm inoculated sets. Our results are in good agreement with these previous findings.

Microbial respiration ($\text{mg/m}^2/\text{hr}$) in control and amended sets has been presented in Table 3. The control set exhibited a decline in respiration from 12.6 \pm 0.81 $\text{mg/m}^2/\text{hr}$ to 9.51 \pm 0.62 $\text{mg/m}^2/\text{hr}$ during the experimental period of 28 days. All the amended sets showed significantly higher ($p < 0.01$) microbial

respiration in comparison to control. The highest value of 27.2 \pm 1.8 $\text{mg/m}^2/\text{hr}$ was observed in F1 on 28th day of incubation. In both the farm yard and poultry manure amended spoil sets higher microbial respiration was observed in earthworm inoculated sets in comparison to the sets without worms. Higher microbial respiration in F1 was likely due to higher bacterial population. Ross and Robert (1970) attributed the variation in the soil microbial respiration to the bacterial load and certain environmental factors such as soil pH, % C etc. Our results corroborate this earlier observation.

ANOVA test results (Table 4) indicated significant difference ($p < 0.01$) in the physicochemical parameters, bacterial population and respiration between treatments. The bacterial population and respiration however did not show significant difference between days of incubation.

CONCLUSION

Analysis of control and amended spoil at different periods of incubation indicated that organic input in iron mine spoil enhance the level of essential soil nutrients and make the spoil a congenial habitat for proliferation of soil microbes which play significant role in maintenance of soil quality. Significantly higher microbial population and respiration in the manure amended sets suggests that the mine spoil can be reclaimed by suitable organic amendments. The reclamation can further be accelerated by utilizing suitable earthworm varieties, which could help in aeration, enhanced microbial activity and nutrient, turn over. Thus microbial colonization through suitable organic amendments will certainly help in reclamation

of the sterile iron mine spoil dumps and facilitate growth of vegetation.

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