

Short communication

Enriching vermicompost by nitrogen fixing and phosphate solubilizing bacteria

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Abstract

The effect of inoculation of vermicompost with nitrogen-fixing *Azotobacter chroococcum* strains, *Azospirillum lipoferum* and the phosphate solubilizing *Pseudomonas striata* on N and P contents of the vermicompost was assessed. Inoculation of N₂ fixing bacteria into vermicompost increased contents of N and P. Enriching vermicompost with rock phosphate improved significantly the available P when inoculated with *P. striata*. During the incubation period, the inoculated bacterial strains proliferated rapidly, fixed N and solubilized added and native phosphate. © 2000 Published by Elsevier Science Ltd.

Keywords: Vermicompost; Enrichment; N–P contents; *Azotobacter chroococcum*; *Azospirillum lipoferum*; *Pseudomonas striata*

1. Introduction

There is a global impact for organic farming through recycling of organic waste for persistent agriculture as well as for a pollution-free environment. For the development of sustainable farming, waste enrichment is of interest. Involvement of earthworms (*Eisenia foetida*) for the degradation of organic wastes and production of vermicompost is near commercialization because loss of nitrogen from agricultural wastes and dung is minimized when the vermicompost is the source of organic matter. There is the possibility of increasing the nitrogen content of compost by inoculation with nitrogen-fixing organisms, and the phosphorus content by the addition of rock phosphate and then inoculation with phosphate-solubilizing bacteria, since direct application of rock phosphate is not useful, particularly in neutral and alkaline soils (Premono et al., 1996). Composting organic refuse with rock phosphate and microbial activities may help to solubilize phosphorus and to increase phosphorus availability to plants. Nitrogen-fixing bacteria, besides fixing N, solubilize P due to production of organic acids and enzymes (Kumar and Narula, 1999). The present experiment was designed to determine how the inoculation of N-fixing and P-solubilizing bacteria

would be able to bring changes in N and P contents of vermicompost.

2. Methods

Charcoal-based nitrogen-fixing and phosphate-solubilizing bacterial cultures were prepared in the laboratory of the Department of Microbiology, CCS Haryana Agricultural University, Hisar. Four nitrogen-fixing bacterial strains: *Azotobacter chroococcum* (Mac27), *A. chroococcum* (54-1), *A. chroococcum* (35-47) and *Azospirillum lipoferum*, with one phosphate-solubilizing bacterium, *Pseudomonas striata*, were used for enrichment studies.

2.1. Preparation of vermicompost

Mature vermicompost (0.2–1 mm size) was used in the experiment, and this was prepared from chopped stalk and leaves of pearl millet (*Pennisetum glaucum*) which had been moistened to 75% and subsequently partially decomposed in pits of (4 ft W × 2.5 ft D × 4 ft L) under anaerobic conditions for three weeks. Similarly cattle dung was partially decomposed under anaerobic conditions. These partially decomposed materials of plant residues and dung were mixed together in the ratio of 3:1 by volume (Plant residues:Dung). This mixture was put in beds (4 ft W × 2 ft H × 5 ft L). The earthworms

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(*Eisenia foetida*), about 1000 per 100 kg material, were introduced into this bed. They were allowed to act for 80 days, which converted the whole material into a fine product named 'vermicompost'. The composition of vermicompost varied with the mineral contents in the agriculture waste used, 1–1.5 N, 0.7–1.1 P and 1.5–2.5 K (g/100 g of dried vermicompost).

2.2. Microbial inoculation of vermicompost

Each experimental treatment consisted of 1 kg of prepared vermicompost, which was placed in a polythene beaker and inoculated with 2.00 g charcoal-based bacterial cultures containing 10^8 cells/g. Water was added to 50% water holding capacity and the whole material thoroughly mixed.

Treatments:

1. Vermicompost + *A. chroococcum* (Mac27)
2. Vermicompost + *A. chroococcum* (54 – 1)
3. Vermicompost + *A. chroococcum* (35 – 47)
4. Vermicompost + *A. lipoferum*
5. Vermicompost + *P. striata*
6. Vermicompost + *P. striata* + 1% Mussoorie rock phosphate (MRP)

Mixtures were incubated for enrichment at 32°C for nine weeks. Bacterial population, nitrogen and available phosphorus contents (g/100 g) were determined at 0, 15, 30, 45, 60 and 75 days. The experiment had three replications for each treatment and data were subjected to statistical analysis. The inoculated bacterial population

was determined using the standard dilution spread-plate method described by Kale et al. (1992). *Azotobacter* count was determined using Jensen medium (Jensen, 1951), *Azospirillum* by Okon et al. (1977) medium and *Pseudomonas* by Pikovskaya (1948). Nitrogen was determined by the method of Jackson (1973), while P was determined by the method of John (1970).

3. Results and discussion

The results presented in Table 1 show that there was a significant increase in the inoculated bacterial populations in vermicompost by the second week. Maximum numbers were found between 45–60 days. After the 60th day there was a decline in count of microbes.

Data of Table 2 show an increase in N and available P contents during the incubation period. Initially at day 0 vermicompost contained only 1.40 (g/100g) of N which was increased to 2.72 (g/100 g) at the 60th day after inoculation with *A. chroococcum* (Mac27). Similarly, with inoculation of other strains of *Azotobacter*, N content increased up to 2.53 and 2.50 (g/100 g). *Azospirillum lipoferum* also increased N content up to 2.18 (g/100 g) but this bacterium was less efficient than *Azotobacter* strains.

The inoculated phosphate-solubilizing bacterium *P. striata*, caused a significant effect on the available P content in vermicompost when inoculated alone or with 1% MRP, but available P content was greater with MRP and *P. striata* combination (1.97) at 60th day.

Table 1
Survival of nitrogen fixing and phosphate solubilizing bacterial strains per g vermicompost

Incubation period (days)	<i>A. chroococcum</i> (Mac27)[$\times 10^3$]	<i>A. chroococcum</i> (54-1)[$\times 10^3$]	<i>A. chroococcum</i> (35-47)[$\times 10^3$]	<i>A. lipoferum</i> [$\times 10^3$]	<i>P. striata</i> [$\times 10^3$]	<i>P. striata</i> +1%MRP[$\times 10^3$]
0	8.9	12.0	11.0	9.0	12.3	12.2
15	19.0	24.2	18.9	25.3	19.1	19.3
30	28.3	36.3	29.0	39.6	30.0	29.6
45	45.0	58.2	57.0	60.3	54.3	52.7
60	51.8	59.8	59.6	63.7	59.0	58.8
75	49.3	58.0	55.8	59.1	53.6	51.3
SEM \pm	0.14	0.12	0.13	0.16	0.17	0.15
C.D. at 5%	0.41	0.34	0.36	0.44	0.48	0.42

Table 2
Enriched nitrogen and available phosphorus contents (g/100 g) in vermicompost

Incubation period	<i>A. chroococcum</i> (Mac27)		<i>A. chroococcum</i> (54-1)		<i>A. chroococcum</i> (35-47)		<i>A. lipoferum</i>		<i>P. striata</i>		<i>P. striata</i> +1%MRP	
	N	P	N	P	N	P	N	P	N	P	N	P
0	1.40	1.10	1.40	1.10	1.40	1.10	1.40	1.10	1.40	1.10	1.40	1.10
15	1.69	1.29	1.76	1.25	1.70	1.31	1.49	1.18	1.50	1.18	1.52	1.43
30	1.74	1.30	1.83	1.35	1.99	1.30	1.63	1.29	1.58	1.20	1.60	1.50
45	2.63	1.39	2.48	1.46	2.43	1.31	2.00	1.36	1.62	1.49	1.60	1.56
60	2.72	1.46	2.53	1.42	2.50	1.39	2.18	1.41	1.68	1.51	1.67	1.97
75	2.73	1.45	2.52	1.42	2.53	1.39	2.16	1.40	1.68	1.52	1.68	1.97
SEM \pm	0.019	0.014	0.017	0.012	0.019	0.011	0.201	0.012	0.018	0.013	0.016	0.015
C.D. at 5%	0.053	0.039	0.048	0.034	0.053	0.031	0.596	0.034	0.048	0.036	0.044	0.042

Without addition of MRP the available P content was 1.51 (g/100 g). At 75th day N and P contents were more or less similar to those of the 60th day.

Addition of rock phosphate inoculated with *P. striata* led to more availability of P, most likely due to the production of organic acids by the bacteria which solubilized the rock phosphate (Premono et al., 1996). The P content in other treatments was higher at 45th and 60th day than at day 0, which was due to release of P present in the agricultural wastes.

It is evident from this experiment that *Azotobacter*, *Azospirillum* and *Pseudomonas* inoculation helped to increase the N and P contents of vermicompost, and rock phosphate was solubilized during composting.

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