International Journal of Recycling of Organic Waste in Agriculture (2021)10: 157-166
Doi: 10.30486/IJROWA.2021.1902019.1087

ORIGINAL RESEARCH

Microbiological integration for qualitative improvement of vermicompost

Sujit Mal[1], G.N. Chattopadhyay[2*], Kalyan Chakrabarti[3]

Received: 12 June 2020 / Accepted: 25 March 2021 / Published online: 30 May 2021

Abstract

Purpose This study was carried out to assess the effect of integrating microbiological fortification with vermicomposting process on biofertilizing quality of vermicompost as well as improvement of the availability of nitrogen and phosphorus in the product.

Method A mixture of cow dung and vegetable market waste (1:1) was used for vermicomposting with *Eisenia foetida* as the decomposer earthworm @ 10 no.of worm kg^{-1} substrate. Nitrogen fixing and phosphate solubilizing bacteria (NFB and PSB)*Azotobacter chroococcum and Pseudomonas fluorescens* were integrated with this composting process at varying doses viz. 0,5 and 10 g kg^{-1} substrate under different combinations. Changes in the population of these two bio-fertilizing microorganisms and the availability of relevant nutrients in the substrates were monitored periodically to assess the behaviors of these microorganisms and their effects on the produced vermicompost.

Results All the inoculations resulted in substantial increments in population of both NFB and PSB over the control. However, the increments were more prominent for NFB than the PSB. Significant increments in the amount of mineralized nitrogen and solubilized phosphate over the control were observed in almost all the treatment combinations.

Conclusion Integration of nitrogen fixing and phosphate solubilizing bacteria with vermicomposting process resulted in substantial enrichment of the product. This benefit was observed not only in terms of increased availability of the two major plant nutrients of concern viz. N and P, but also in significant improvement in the population of the inoculated microorganisms turning the product into a potential source of bio-fertilizers.

Keywords Vermicomposting, Microbial fortification, Compost quality, Nutrient availability, Bio-fertilizer population

Introduction

In recent years, vermicomposting biotechnology has emerged as an efficient method for recycling varying kinds of organic wastes in the form of compost with the help of gut microorganisms of epigeic earthworms (Singh et al. 2011; Bhat et al. 2017; Mupanwa and Mnkeni 2018). The nutrient status of vermicompost is generally higher than the traditional composts (Chattopadhyay 2012). Yet, any effort for further improvement of the nutrient content as well as the microbial quality of this compost will help to increase its efficiency in upgrading the soil health. Several investigations have been made to increase the nutrient content of vermicompost though addition of mineral fertilizers (Bhattacharya and Kumar 2009). Adhami et al. (2014) observed vermicomposting to increase P solubilization from rock phosphate and attributed this behavior to release of organic acids during the decomposition process. Similar results have been reported by workers like Unuofin et al. 2014; Mihreteab et al. 2016 and others. The crucial link between earthworms and microbes during the vermicomposting process has also prompted several researchers in trying to modify the microbiological composition of vermicomposts through inoculation of

✉ G.N.Chattopadhyay
gunin.c2010@gmail.com

1 College of Agriculture, Susunia (Extended Campus of Bidhan Chandra Krishi Viswavidyalaya),Chhatna, West Bengal, India
2 Seacom Skills University,Kendradangal, West Bengal, India
3 Ex-Institute of Agricultural Science, Calcutta University, 35 Ballygunj Circular Road, Kolkata, West Bengal, India

some specialized microbial cocktails (Mal et al. 2013; Mupambwa and Mnkeni 2018).This approach has led to number of studies to strengthen the microbiological properties of vermicompost (Mupambwa et al. 2016). However, in spite of all these efforts, information on microbiological management with regard to nutrient enrichment of vermicompost has remained limited. Primarily, vermicomposting is an earthworm gut microbe induced organic waste degradation system. Therefore, adoption of some suitable management practice with the help of bio-fertilizing microbes will be particularly useful for biologically induced qualitative improvement of vermicompost as a rich source of not only the relevant nutrients but also the inoculated bio-fertilizers. Gaur (2006), while emphasizing the need for improvement of the quality of traditional composts, suggested inoculation of nitrogen fixing bacteria (NFB) for increasing the nitrogen content and phosphate solubilizing microorganisms (PSM) for improving the availability of phosphorus in the composts. In addition, NFB inoculation can also hasten the rate of organic waste decomposition by narrowing the C: N ratio (Manna et al. 2012). On the other hand, better P nutrition generated by PSM is likely to enhance the growth and activity of general microbial population in the substrates. Adoption of any such bio-fertilizing microbial management practice for vermicomposting will , therefore, help to sustain the soil health not only through the supply of the nutrient rich organic matter (Busato et al. 2012) but also by the consequent multiplication of these bio-fertilizing microbes to the soils. However, information pertaining to such microbiological manipulation with regard to enhancement in nutrient availability as well as bio-fertilizer supplying capacity of vermicompost is limited. Under this context, the possibility of improving the nutritional as well as bio-fertilizing quality of vermicompost through microbiological fortification has been attempted in the present investigation. Nitrogen fixing bacteria (NFB), *Azotobacter chroococcum* and phosphate solubilizing bacteria (PSB) *Pseudomonas fluorescens* were used for the study as the biofertilizing inoculums. Of these, *Azotobacter chroococcum* has been described as an efficient NFB for enriching various composting processes (Manna et al. 2012). On the other hand, *Pseudomonas fluorescens* is a common PSB which is having both phosphate solubilizing as well as bio-controlling properties (Kalayu 2019; Panpatte et al. 2016). Both of these microbial inoculants are commonly available in the market and may be procured easily by the compost producers. It has been hoped that adoption of this simple microbial management practice will be highly effective to improve the nutritional as well as bio-fertilizing capacity of vermicompost; thus, enabling this organic input to perform more beneficial roles in betterment of soil health through increased availability of the major plant nutrients like N and P and also the bio-fertilizing microorganisms.

Materials and methods

The study was carried out under yard condition using earthen pots tapering towards lower end having 25 cm diameter at the top and 13 cm at lower end with a height of 20 cm and perforation in the bottom to avoid water accumulation. Organic wastes containing mixture of cow dung and vegetable market waste at 1:1 ratio (w/w) were taken in 21 nos. of such pots @ 1kg waste per pot. The substrates were maintained under moist (40-50%) condition through periodic addition of water. Cessation of the initial thermophilic condition, i.e., reduction of temperature below 40 ^{0}C, in all the treatments took 9-11 days which brought down the substrate temperatures to 36--38^{0}C. At this stage, two common nitrogen fixing and phosphate solubilizing bacteria, *Azotobacter chroococcum* and *Pseudomonas fluorescens* were inoculated in the vermicomposting process under different combinations using six treatments. Details of the treatments along with the doses of microbial inoculation and the concentrations of the inoculated organisms have been provided in Table 1. In addition, the study included a control (T_1) which received only organic wastes and earthworms, without any microbial treatment. Thus, in total, seven treatments each with three replicates were used for this study. After 3 days of inoculation of the microorganisms, epigeic earthworm *Eisenia foetida*, a common decomposer earthworm used for vermicomposting (Ravindran et al. 2015; Usmani et al. 2017) ,was released to each pot @ 10 no.of worm kg^{-1} waste. This number has been reported to be within optimum stocking range of earthworms for vermicomposting by workers like Sahariah et al. (2014), Das et al. (2016) and others. The substrate temperatures during the vermicomposting period of 60 days ranged between 31 and 38 ^{0}C under different treatments exhibiting a congenial environment for microbial activity as well as vermicomposting.

Composite samples were drawn from the substrates in each of the pots at 15 day intervals from the date of releasing the earthworms and were analyzed for NH_4^+ and NO_3^- nitrogen with the help of a Kjeltec distillation unit. The alkaline distillation method described by Jackson (1973) without and with Davarda's alloy was used for estimation of these two forms of N. Availability of phosphorus was determined by Olsen's method using 0.5M $NaHCO_3$ as the extractant (Jackson 1973) with the help of a spectrophotometer. Populations of nitrogen fixing bacteria (NFB) and phosphate solubilizing bacteria (PSB) were estimated after 60 days of incubation. NFB was estimated by using N free Jenson's agar medium. For enumeration of PSB, Pikovskaia's tricalcium phosphate agar medium was used. In each case, enumeration was done by counting the colony forming units using serial dilution technique as described by Johnson et al. (1956).

A portion of the substrate from each of the replicates was weighed under both moist and air dried conditions during each of the sampling and the results of relevant analyses were corrected for air dry condition. Moisture status of the composting systems were also maintained based on these observations.

Table 1 Treatment combinations used in the study

Treatments	Microbial combinations (g kg^{-1})
T_1 (control)	Organic waste (OW) + Earthworm (EW)
T_2	OW + 10g PSB+ EW
T_3	OW +5g PSB + 5g NFB + EW
T_4	OW + 10g PSB + 5g NFB + EW
T_5	OW + 10g PSB + 10g NFB + EW
T_6	OW + 10g NFB + EW
T_7	OW + 5g PSB + 10g NFB + EW

Earthworm was added @ 10 no. kg^{-1} waste
NFB and PSB were procured from market at a concentration of 10 x 10^6 no. g^{-1} for each.

Results and discussion

Colony forming units (CFU) of nitrogen fixing bacteria (NFB) in the substrates under different treatments increased gradually with the progress of vermicomposting (Table 2). As discussed by Wallwork (1984) and then by other workers (Bhattacharya and Chattopadhyay 2005; Gomez-Brandon and Dominguez 2014), micro flora present in the organic wastes are consumed by the earthworms along with the food materials and are provided congenial environment in the worm guts to grow profusely. These microorganisms are subsequently released to the vermicomposting system through the earthworm excreta, thus increasing their population in the substrates. This activity helped to maintain an almost consistently increasing trend in the occurrence of NFB in the vermicomposts under the present study also (Table 2).The increments were comparatively slower during the initial phases of composting but gained pace with the progress of the study, as more nutrients were released in available forms. At the end of the vermicomposting period, all the treatments with NFB resulted in statistically significant increase in the population of this microflora over the control. Similar increase in the NFB population during the course of vermicomposting has been reported by Bhattacharya and Chattopadhyay (2005) also. The mean population of NFB in the substrates under different treatments have been presented in Fig. 1. The Fig. 1 indicates the trend of general occurrence of this microbe under different treatments during the span of vermicomposting. As shown in the Fig. 1, microbial inoculations resulted in 19.1 to 65.3% increment in mean population of NFB over the control. It was interesting to observe that simultaneous inoculation of PSB with NFB in different combinations helped to maintain increased populations of NFB in the substrate. After completion of vermicomposting, the highest occurrence of NFB (58.66x10^6 no.g^{-1}) was found in the treatment receiving combined inoculation of NFB and PSB @ 10g kg^{-1} each. This value was statistically at par with the corresponding NFB concentrations in T6 and T_3 treatments bearing sole use of NFB @10g kg^{-1} and combination of NFB and PSB @5 g kg^{-1}, respectively .The NFB population in the treatment T_2, with only PSB @ 10g kg^{-1} and no NFB, also increased significantly over the control. This synergistic effect of PSB on NFB population may be due to increase in availability of phosphorus in the PSB treated substrates. P plays a critical role in cell division (Weil and Brady 2018). Hence, these microorganisms, which propagate through cell division, depend largely on the availability of P for their multiplication .Therefore, in the present study, better P availability under PSB treatments helped in more rapid multiplication of NFB, too. Such proliferation of NFB under co-inoculation of phosphate solubilizing microorganisms has been discussed by Subba Rao (1999) also. This increased population of NFB not only improves the quality of the compost by fixing atmospheric N_2 into the substrate but also tends to hasten the composting process by narrowing the C: N ratio (Gaur 2006). In addition, the high NFB concentrations in these NFB treated vermicomposts indicate that such vermicompost may act as a rich source of nitrogen fixing bio-fertilizer after its application to the soil.

Availability of NH_4^+ nitrogen in the substrates increased more or less consistently with the period of incubation in all the treatments (Table 3). This behavior was attributed to gradual mineralization of the organic wastes releasing nitrogen to NH_4^+ form. The differences among the treatments were narrower during the initial phase of the study but widened as the vermicomposting process proceeded further. It was interesting to observe that although T_5 treatment showed the highest mean CFU of NFB, yet the amount of NH_4^+ nitrogen was not that high in this treatment. This behavior was attributed to large scale nitrification of the ammonified nitrogen under the prevailing aerobic condition of the vermicomposting process. As will be discussed later, this treatment showed highest occurrence of NO_3^- nitrogen (Table 4).

Values of NO_3^- nitrogen also increased on incubation (Table 4) owing to gradual nitrification of the NH_4^+ nitrogen in the substrates. Nitrifying bacteria are essentially aerobic in nature (Tilak et al. 2010) and ver-

Table 2 C.F.U of nitrogen fixing bacteria (x × 10^6 g^{-1}) in different treatments during the period of incubation

Treatments	C.F.U. at 15 days interval	C.F.U at 30 days interval	C.F.U at 45 days interval	C.F.U at 60 days interval
T_1 = Control (O.W)	18.66	21.33	22.0	30.33
T_2 = 10g P.S.B. + O.W + E.W	20.66	25.33	24.66	39.33
T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W	24.66	26.00	24.66	51.33
T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W	21.33	27.33	24.66	38.0
T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W	26.66	31.33	36.0	58.66
T_6 = 10 g N.F.B+ O.W+ E.W	22.00	24.33	26.00	53.66
T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W	20.66	26.66	46.66	51.33
.C.D	**6.46**	**5.88**	**5.91**	**8.17**

Table 3 Availability of NH_4-N (mg kg^{-1}) in different treatments during the period of incubation

Treatments	NH_4-N at 15 days interval	NH_4-N at 30 days interval	NH_4-N at 45 days interval	NH_4-N at 60 days interval
T_1 = Control (O.W)	305.82	412.96	448.18	688.62
T_2 = 10g P.S.B. + O.W + E.W	428.89	759.75	764.91	890.99
T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W	566.52	448.27	1055.68	1055.65
T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W	453.52	537.65	606.28	551.54
T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W	595.45	782.23	882.23	887.63
T_6 = 10 g N.F.B+ O.W+ E.W	538.15	559.79	892.32	1056.04
T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W	532.14	551.64	532.27	924.24
C.D.	**62.54**	**49.12**	**69.33**	**80.42**

Table 4 Availability of NO_3-N (mg kg^{-1}) in different treatments during the period of incubation

Treatments	NO_3-N at 15 days interval	NO_3-N at 30 days interval	NO_3-N at 45 days interval	NO_3-N at 60 days interval
T_1 = Control (O.W)	448.16	504.20	489.27	504.26
T_2 = 10g P.S.B. + O.W + E.W	477.94	826.13	726.28	830.18
T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W	473.18	766.87	747.55	1146.27
T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W	478.51	532.20	532.26	932.84
T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W	586.28	517.88	510.89	1148.21
T_6 = 10 g N.F.B+ O.W+ E.W	495.30	1089.09	922.30	1055.61
T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W	522.17	536.21	597.14	987.60
C.D.	**51.14**	**56.63**	**48.17**	**61.97**

micomposting is an aerobic decomposition process (Chattopadhyay 2005). Hence, the nitrifying microbes could work efficiently under this congenial environment of vermicomposting and increased the amount of NO_3^- form of nitrogen in the system.

Since both NH_4^+ and NO_3^- N constitute the readily available forms of this nutrient, the total availability of N as $NH_4^+ + NO_3^-$ has been presented in Fig. 2. The treatments exhibiting higher occurrence of NFB, in general, showed higher concentrations of mineralized ($NH_4^+ + NO_3^-$) forms of nitrogen. This was attributed to hastening of decomposition owing to lowering in carbon: nitrogen ratio by NFBs (Raj and Antil 2011; Ravindran and Mnkeni 2016). Such accelerated mineralization of organic materials helped to release more amount of nitrogen to mineralized forms.

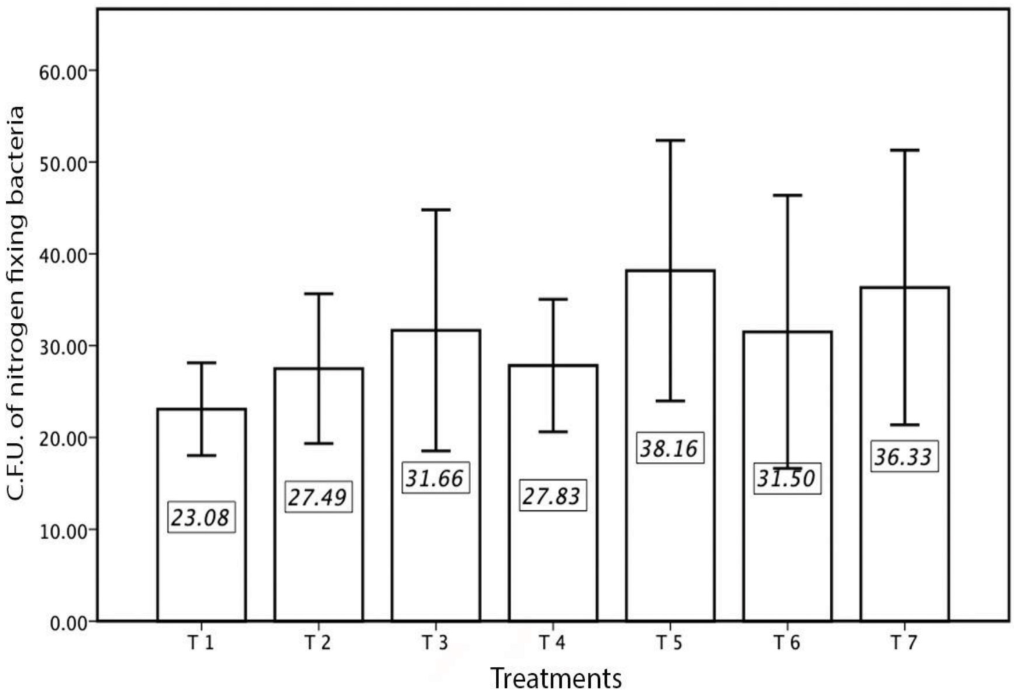

Fig. 1 Mean occurrence of NFB in the substrates under different treatments

T_1 = Control :Organic waste (OW) + Earthworm (EW); T_2 =10g P.S.B. + O.W + E.W; T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W; T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W; T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W; T_6 = 10 g N.F.B+ O.W+ E.W; T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W.

Occurrences of phosphate solubilizing bacteria (PSB) in different treatments during the period of vermicomposting have been presented in Table 5.The control treatment showed the lowest values of PSB during the entire period of incubation due to obvious reasons. On the other hand, inoculation of PSB helped to increase its occurrence in other treatments. Among different treatments with PSB, inoculation @ 10g kg^{-1} substrate (T_2) showed the highest and statistically superior population of PSB ($18.33x10^6$ no. g^{-1}). On the other hand, use of same dose of PSB along with different concentrations of NFB (T_4, T5) resulted in marginal decline in PSB values, as compared to T_2 treatment. This behavior was not in conformity to what was observed in case of occurrence of NFB in presence of PSB in this study (Table 2). While NFB was benefited by the phosphorus released by PSB from the organic wastes during vermicomposting (Subba Rao 1999), the PSB did not receive such mutual benefit from NFB. In the former case, the PSB induced higher availability of P acted as a booster to the multiplication of unicellular NFB. On the other hand, mineralization of N in all the vermicomposted materials resulted in release of sufficient N in available form for the growth of the microbial population irrespective of the effects of NFB in fixing atmospheric nitrogen (Fig. 2). Hence, co-inoculation of NFB did not provide any additional benefit to the PSB population with regard to their N nutrition, rather the PSB population had to compete with the NFB for their growth.

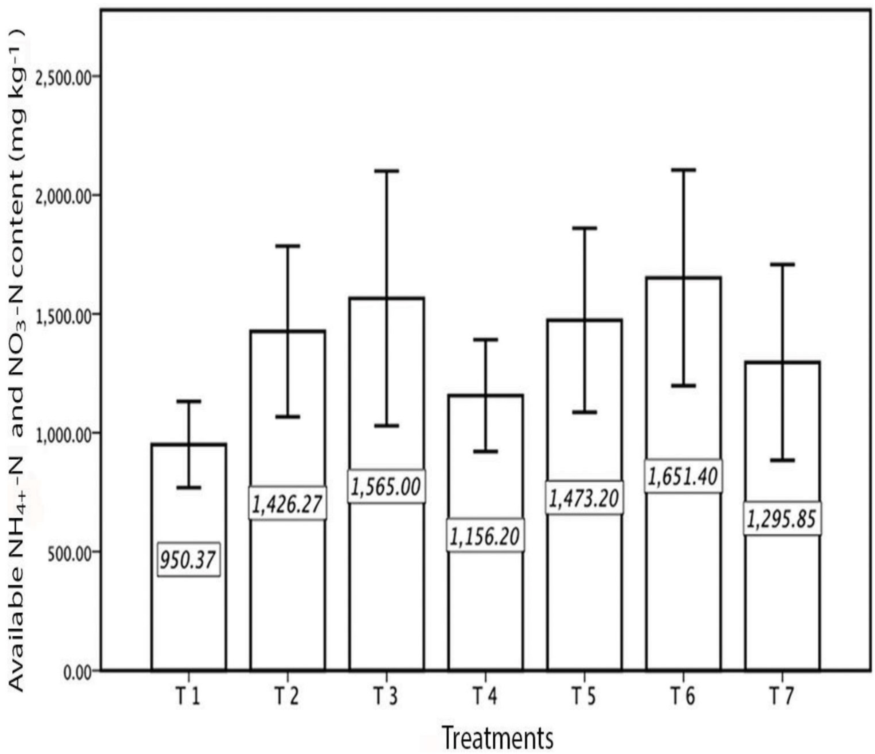

Fig. 2 Mean NH_4^+ and NO_3^- -N content (mg kg^{-1}) in the substrates under different treatments

T_1 = Control: Organic waste (OW) + Earthworm (EW); T_2 =10g P.S.B. + O.W + E.W; T_3= 5g P.S.B. + 5 g N.F.B+ O.W+ E.W; T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W; T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W; T_6= 10 g N.F.B+ O.W+ E.W; T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W.

Availability of phosphorus in the substrates under different treatments have been presented in Table 6. Inoculation of the biofertilizers helped to increase the availability of phosphorus significantly over the control in all the treatments. Among them, use of higher dose of PSB (10g kg^{-1}) in combination with NFB resulted in comparatively higher availability of phosphorus than the treatment with same dose of PSB alone (T_2). While highest availability of phosphorus (996.31 mg kg^{-1}) was observed in the treatment with both PSB and NFB @ 10g kg^{-1} each (T_5), the next highest value of available phosphorus was found in the treatment with PSB @ 10g and NFB @ 5g kg^{-1} substrate. This may be due to larger transformation of organic phosphorus to mineral forms due to increased rate of decomposition effected by narrowing of C: N ratios in presence of NFBs (Gaur 2006). In addition to phosphate solubilizing microorganisms, several nitrogen fixing bacteria can also solubilize phosphorus from insoluble sources (Subba Rao 1999). This behavior probably resulted in synergistic effect on phosphorus solubilization when both PSB and NFB were inoculated to the vermicomposting system. This hypothesis may be supported by the observation that use of NFB alone @ 10g kg^{-1} substrate (T_6) also resulted in statistically significant increment in available phosphorus status over control (Table 6).

Table 5 C.F.U of Phosphate solubilizing bacteria (P.S.B.) in different treatments during the period of incubation: ($x \times 10^6$ g^{-1})

Treatments	C.F.U at 15 days interval	C.F.U at 30 days interval	C.F.U at 45 days interval	C.F.U at 60 days interval
T_1 = Control (O.W)	7.66	8.33	8.33	10.33
T_2 = 10g P.S.B. + O.W + E.W	10.66	12.66	14.00	18.33
T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W	8.0	11.0	10.66	12.33
T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W	9.66	11.33	10.66	13.0
T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W	9.66	10.66	10.66	14.33
T_6 = 10 g N.F.B+ O.W+ E.W	8.0	10.33	9.0	10.66
T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W	8.66	8.66	10.0	0. 12
C.D.	**3.44**	**3.01**	**2.00**	**3.45**

Table 6 Availability of P_2O_5 (mg kg^{-1}) in different treatments during the period of incubation

Treatments	P_2O_5 at 15 days interval	P_2O_5 at 30 days interval	P_2O_5 at 45 days interval	P_2O_5 at 60 days interval
T_1 = Control (O.W)	331.19	320.62	348.72	564.14
T_2 = 10g P.S.B. + O.W + E.W	455.81	474.08	523.31	638.45
T_3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W	546.18	587.16	567.21	783.06
T_4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W	460.53	455.5	517.52	872.29
T_5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W	593.69	601.53	577.63	996.31
T_6 = 10 g N.F.B+ O.W+ E.W	417.83	422.70	511.89	643.48
T_7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W	407.18	448.71	481.46	631.50
C.D.	**38.89**	**45.02**	**44.96**	**63.32**

Comprehensive contributions of different biofertilizer treatments on resultant populations of NFB and PSB and also the increments in available N and P_2O_5 status in the biologically fortified vermicompost were taken into account by considering the overall benefits achieved by each of the treatments. Several of these treatments showed appreciable improvements with regard to the biofertilizing capacity and also the availability of relevant plant nutrients. Among them, the combination of NFB and PSB each @ 10 g inoculum kg^{-1} of substrate showed better overall performance. This treatment resulted in the highest occurrence of NFB (Fig. 1) and 2nd highest concentrations of PSB (Fig 3). The availability of P_2O_5 was the highest in this treatment while occurrence of total mineralized N (NH_4^+ + NO_3^-) was also among the top three.

Such value addition in the vermicompost in term of biofertilizer fortification is likely to fetch better economic return also. With integration of 10 g inoculum of NFB and PSB each, this microbe enriched vermicompost (T_5) resulted in a final product carrying 58.66 x10^6CFU of NFB and 14.35x 10^6CFU of PSB making the product a rich source of both nitrogenous and phosphatic biofertilizing microorganisms. Hence, this biologically fortified vermicompost will serve not only as an organic manure with better N and P supplying capacity but also as a potential source of two important biofertilizers; thus, minimizing the necessity of separate application of N and P biofertilizers in soils.

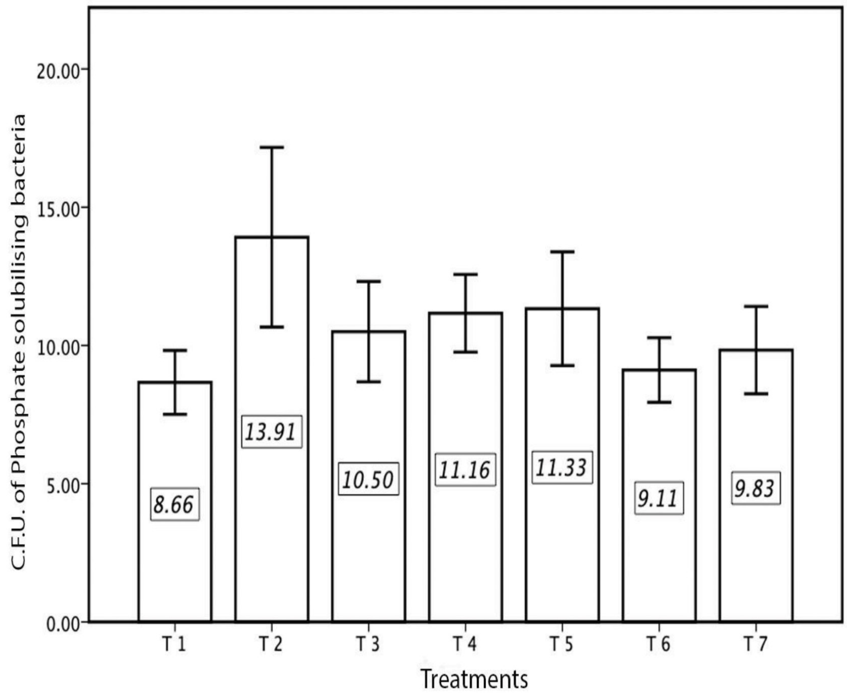

Fig. 3 Mean occurrence of PSB in the substrates under different treatments

T1 = Control :Organic waste (OW) + Earthworm (EW); T2 =10g P.S.B. + O.W + E.W; T3 = 5g P.S.B. + 5 g N.F.B+ O.W+ E.W; T4 = 10g P.S.B. + 5 g N.F.B+ O.W+ E.W; T5 = 10g P.S.B. + 10 g N.F.B+ O.W+ E.W; T6 = 10 g N.F.B+ O.W+ E.W; T7 = 5g P.S.B. + 10 g N.F.B+ O.W+ E.W.

Conclusion

The study dealt with the effects of integrating nitrogen fixing and phosphate solubilizing bacteria (NFB and PSB) with vermicomposting process on availability of N and P and also the biofertlizer supplying capacity of this microbiologically fortified vermicompost. The results of inoculation of different combinations of *Azotobacter chroococcum and Pseudomonas fluorescens* in the vermicomposting process of cow dung and vegetable market waste mixture using *Eisenia foetida* as the decomposer earthworm was assessed in this investigation. The study showed significant increase in the population of these two bio-fertilizing microbes over the control depending on the doses of their inoculation and combination. Between the two, however, the increment was more prominent for the NFB than the PSB. Apparently, PSB provided increased sustenance to NFB population through better P nutrition. Enhanced occurrences of these bio-fertilizing microbes resulted in improved availability of the relevant nutrients viz. N and P in the resultant vermicomposting. Significant increments in the amount of mineralized nitrogen and solubilized phosphate over the control were recorded in almost all the treatment combinations, of which combined use of NFB and PSB @ 10g kg^{-1} each appeared to be the most effective.

The study showed integration of nitrogen fixing and phosphate solubilizing bacteria with vermicomposting process to improve the availability of these two nutrients in the product. In addition, this biologically fortified vermicompost appeared to act as a rich source of N and P biofertilizing microbes, thus minimizing the necessity of separate application of these biofertilizers.

Compliance with ethical standards

Conflict of interest The authors declare that there are no conflicts of interest associated with this study.

References

Adhami E, Hosseini S, Owliaie H (2014) Forms of phosphorus of vermicompost produced from leaf compost and sheep dung enriched with rock phosphate. Int J Recycl Org Waste Agricult 3: 68. https://doi.org/10.1007/s40093-014-0068-9

Bhat SA, Singh J, Vig AP (2017) Earthworms as organic waste managers and biofertilizer producers. Waste Biomass Valor 9: 1073–1086. https://doi.org/10.1007/s12649-017-9899-8

Bhattacharya SS, Chattopadhyay GN (2002) Increasing bioavailability of phosphorus from fly ash through vermicomposting. J Environ Qual 31: 2116-2119

Bhattacharya SS, Chattopadhyay GN (2004) Transformation of nitrogen during vermicomposting in fly ash. Waste Manag Res 22(6): 488-491

Bhattacharya P, Kumar D (2009) Enrichment of vermicompost with urea, rock phosphate, azolla, pressmud and biofertilisers. Agrobios Newsletter (July). pp- 39-40

Busato JG, Lima LS, Aguiar NO, Canellas LP, Olivares FL (2012) Changes in labile phosphorus forms during maturation of vermicompost enriched with phosphorus-solubilizing and diazotrophic bacteria. Bioresour Technol 110: 390–395. https://doi.org/10.1016/j.biortech.2012.01.126

Chattopadhyay GN (2005) Vermicomposting as a biotechnological tool for recycling organic wastes. In: Ghosh TK, Chakrabarti T, Tripathi G (eds) Biotechnology in Environmental Management 1: 135-145

Chattopadhyay GN (2012) Use of vermicomposting biotechnology in agriculture. Int J Recycl Org Waste Agricult 1: 1-8. https://doi.org/10.1186/2251-7715-1-8

Das D, Bhattacharyya P, Ghosh BC, Banik P (2016) Bioconversion and biodynamics of *Eisenia foetida* in different organic wastes through microbially enriched vermiconversion technologies. Ecol Eng 86: 154–161. https://doi.org/10.1016/j.ecoleng.2015.11.012

Gaur AC (2006) Biofertilizers in sustainable agriculture. ICAR, New Delhi

Gomez-Brandon M, Dominguez J (2014) Recycling of solid organic wastes through vermicomposting: microbial community changes throughout the process and use of vermicompost as a soil amendment. Crit Rev Environ Sci Technol 44(12): 1289–1312. https://doi. org/10.1080/10643389.2013.763588

Jackson ML (1973) Soil chemical analysis. Prentice Hall of India Pvt Ltd, New Delhi

Johnson LF, Curl EA, Bond JS, Friboury HA (1956) Methods for studying soil microflora plant disease relationships. Burgers Publication Comp., Minnepolis, Minnesota

Kalayu G (2019) Phosphate solubilizing microorganisms: Promising approach as biofertilizers. Int J Agron. Article ID. 4917256. https://doi.org/10.1155/2019/4917256

Mal S, Chattopadhyay GN, Chakraborti K (2013) Use of some indigenous inputs for improving microbiological activity in vermicomposting system. The Multidiscipl Nat J Carmelight 10 (1): 13-21

Manna MC, Subba Rao A, Sahu A, Singh UB (2012) Compost handbook: Research-production-application. FDCO, New Delhi

Mihreteab HT, Ceglie FG, Aly A, Tittarelli F (2016) Rock phosphate enriched compost as a growth media component for organic tomato (*Solanum lycopersicum* L.) seedlings production. Biol Agric Hortic 32(1): 7–20. https://doi.org/10.1080/01448765.2015.1016114

Mupambwa HA, Mnkeni PNS (2018) Optimizing the vermicomposting of organic wastes amended with inorganic materials for production of nutrient-rich organic fertilizers: A review. Environ Sci Pollut Res. https://doi.org/10.1007/11356-018-1328-4

Mupambwa HA, Ravindran B, Mnkeni PNS (2016) Potential of effective micro-organisms and *Eisenia fetida* in enhancing vermi-degradation and nutrient release of fly ash incorporated into cow dung–paper waste mixture. Waste Manag 48: 165–173. https://doi.org/10.1080/10643389.2013.763588

Panpatte D, Jhala YK, Shelat H, Vyas RV (2016) *Pseudomonas fluorescens:* A promising biocontrol agent and PGPR for sustainable agriculture. In: Singh DP et al (eds) Microbial inoculants in sustainable agricultural productivity. Springer, India: 257-270. https://doi.org/10.1007/978-81-322-2647-5_15

Raj D, Antil RS (2011) Evaluation of maturity and stability parameters of composts prepared from agro-industrial wastes. Bioresour Technol 102(3): 2868–2873. https://doi.org/10.1016/j.biortech.2010.10.077

Ravindran B, Contreras-Ramos SM, Sekaran G (2015) Changes in earthworm gut associated enzymes and microbial diversity on the treatment of fermented tannery waste using epigeic earthworm *Eudrilus eugeniae*. Ecol Eng 74: 394–401. https://doi.org/10.1016/j.ecoleng

Ravindran B, Mnkeni PNS (2016) Bio-optimization of the carbon to nitrogen ratio for efficient vermicomposting of chicken manure and waste paper using *Eisenia fetida*. Environ Sci Pollut Res 23(17): 16965–16976. https://doi.org/10.1007/s11356-016-6873-0

Sahariah B, Sinha I, Sharma P, Goswami L, Bhattacharyya P, Gogoi N, Bhattacharya SS (2014) Efficacy of bioconversion of paper mill bamboo sludge and lime waste by composting and vermiconversion technologies. Chemosphere 109: 77–83

Singh RP, Singh P, Araujo ASF, Ibrahim MH, Sulaiman O (2011) Management of urban solid waste: Vermicomposting a sustainable option. Resour Conserv Recycl 55(7): 719–729. https://doi.org/10.1016/j.resconrec.2011.02.005

Subba Rao NS (1999) Soil microbiology (4thed of soil microorganisms and plant growth). Oxford IBH, New Delhi

Tilak KVBR, Pal KK, De R (2010) Microbes for sustainable agriculture. I.K. Int Publ. House Pvt. Ltd. New Delhi

Unuofin FO, Mnkeni PNS (2014) Optimization of *Eisenia fetida* stocking density for the bioconversion of rock phosphate

enriched cow dung–waste paper mixtures. Waste Manage 34(11): 2000–2006.
https://doi.org/10.1016/j.wasman.2014.05.018

Usmani Z, Kumar V, Mritunjay SK (2017) Vermicomposting of coal fly ash using epigeic and epi-endogeic earthworm species: Nutrient dynamics and metal remediation. RSC Adv 7: 4876

Wallwork JA (1984) Earthworm biology 1st Indian Edn. Gulab Vazirani for Arnold Heinemann. New Delhi

Weil RR, Brady Nyle C (2018) The nature and properties of soils. Global Ed. Pearson Education Ltd. London