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Effect of organic and inorganic additives on some chemical properties of vermicompost, earthworm's biomass and reproduction

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ABSTRACT-The enrichment of bedding for vermicomposting systems with organic and inorganic additives is a technique which can promote the quality of produced vermicompost. Phosphate rock and fishmeal are two available and low-cost substances which can be provided easily and inoculation of vermicompost with phosphate solubilizing microorganisms (PSMs) can enhance the availability of their nutrients. In this research, the effect of using phosphate solubilizing bacteria (PSB) (*Pseudomonas fluorescens*) by adding phosphate rock (0 and 1% W/W) and fishmeal (0 and 1% W/W) on some chemical and biological properties of vermicompost was studied under greenhouse conditions with a factorial arrangement in a completely randomized design with three replications. Phosphate rock application and PSM inoculation decreased N concentration while fishmeal increased it. P concentration increased by adding phosphate rock, fishmeal or PSB or their co-applications. Fishmeal increased micro-nutrients concentration except for Fe. Phosphate rock increased Fe concentration and decreased other micro-nutrients concentration. Electrical conductivity (EC) increased by adding phosphate rock but decreased by fishmeal application. Phosphate rock and fishmeal increased pH. Phosphate rock and PSB decreased the number and weight of earthworms and the number of cocoons. Fishmeal application increased the number and weight of earthworms and number of cocoons. Results indicated that enrichment of vermicompost bedding by additives could increase some essential nutrients and change the growth and biomass of earthworms.

INTRODUCTION

Increasing population has become a concern around the world nowadays. Food health, environmental safety, and soil conservation are some issues that lead human to use sustainable agriculture. Humans' needs have been provided with conserving resources and environment in sustainable agriculture (Lozcano and Dominguez, 2011). Using an organic fertilizer such as vermicompost instead of chemical fertilizers is one of the most important bases in sustainable agriculture (Tilman et al., 2002).

Vermicompost is known as an organic fertilizer that is rich in essential nutrients and improves soil physical and chemical properties (Dominguez and Edwards, 2004). Acting some earthworm species (such as *Eiseniafetida, Lumbricusrubellus* and *Eudriluseugenae)* on many organic matters and residues produces a material which can significantly increase plant growth (Chaudhuri et al., 2000; Manyuchi and Phiri, 2013). Using vermicompost in agriculture can increase plant yield, height, the number of flowers and leaves and plant health because of a large amount of available nutrients, high level of humic acid and other plant growth regulators (Theunissen et al., 2010).

Enriching vermicompost is a technique for improving vermicompost quality and quantity which can be performed by bio-wastes, chemical fertilizers and beneficial microorganisms inoculation such as plant growth promoting rhizobacteria (PGPR) and phosphate solubilizing microorganisms (PSMs) (Mahantaa et al., 2012). Enriched vermicompost with higher nutrient levels can be used in lower rates for agricultural products and to save costs.

Fishmeal is a rich substance used in livestock and fish diets. Fishmeal can be used in agriculture as a useful fertilizer. It has high levels of proteins, lipids, vitamins, nutrients and low levels of carbohydrates (Miles and Chapman, 2006). Phosphate rock is the first material for making phosphate fertilizers, and it is one of the best alternatives in countries that do not have enough materials to produce phosphate fertilizers (Van kauwenbergh, 2001; Haynes, 1984). Researchers showed that applying phosphate rock directly is not very useful. So, it may commonly be used with inoculation of PSMs which can solubilize insoluble phosphate and increase the available forms (Premono et al., 1996; Kumar and Narula, 1999; Kumar and Singh, 2001). Adding insoluble phosphate with PSMs to the bedding materials in vermicompost production can increase available phosphorus in vermicompost. The phosphate solubilizing mechanisms of these microorganisms can be through the production of phosphatases enzymes and organic acids with low molecular weight like gluconic acid, releasing H^+ and chelating compounds (Goldstein, 1986; Kim et al., 1998; Busato et al., 2012).

Additives materials in the vermicomposting process may affect the growth and biomass of earthworm. It is very important to optimize the growth conditions and earthworms reproduction to achieve high efficiency in vermicompost production and vermiculture. Ebadi et al. (2007) showed that tomato, sawdust, sugar–cane, cow manure and soil mixtures produced the best environment for growth and reproduction of earthworms. Mirbolook et al. (2011) showed that sugar beet molasses increased the number of adult and juvenile earthworms, cocoon and biomass. Alikhani et al. (2015) indicated that the most significant earthworm biomass was in treatments of 50 v/v tomato and sugarcane wastes application. Soil application in bed materials increased the number of cocoons and biomass significantly. They reported that the highest amount of earthworm biomass was obtained during the seventh and eighth weeks of experiment and decreased afterward. However, Haimi and Huhta (1986) reported that sewage sludge decreased reproduction (number of cocoons) of earthworms.

So this research was conducted to assess the effect of the enrichment of vermicompost bedding with phosphate rock, fishmeal and phosphate solubilizing bacteria (PSB) on some chemical and biological properties of vermicompost under greenhouse conditions.

MATERIALS AND METHODS

The experiment was carried out under greenhouse conditions, with day/night temperature of 24±3ºC/15±3ºC, 60-70% average relative humidity (RH) and a photoperiod of 14 (h) with photosynthetic photon flux (PPF) of 800 µmol m^{-2} s^{-1}. It was conducted with the factorial arrangement in a completely randomized design with three replications. Treatments consisted of two levels of fishmeal (0 (FM_0) and 1% (FM_1) W/W), two levels of phosphate rock (0 (PR_0) and 1% (PR_1) W/W), and two levels of bacterial treatment (non-bacterial inoculation (B_0) and inoculation with *Pseudomonas fluorescens* (B_1)). The fishmeal was obtained from the Beyza 21 Feed Mill Company. Rotted cow manure was collected from the Animal Research Station of Shiraz University and used as the bedding material for vermicompost production. Air dried cow manure and fishmeal samples were passed through a 2-mm sieve and mixed uniformly. Some chemical properties of fishmeal and rotted cow manure are presented in Table 1. The phosphate rock was provided from Esfordi phosphate mineral complex, Yazd, Iran. Some chemical properties of used phosphate rock are presented in Table 2 based on factory recommendation. *Pseudomonas fluorescens* bacterium with phosphate solubilizing and siderophore production ability was obtained from Soil Biology Lab, Department of Soil Science and Engineering, University of Tehran. The pure bacterial culture was grown on nutrient broth medium in a shaker incubator, at 28°C for 48h (Malekzadeh et al., 2012). Inorganic and organic phosphate-solubilizing ability of the bacterium was measured semi-quantitatively based on the appearance of clear zones (halos) surrounding the bacterial colonies on Sperber solid medium containing $Ca_3(PO_4)_2$ and inositol hexa phosphate as source of inorganic and organic P, respectively (Sperber, 1958; Zarei et al., 2006).

Plastic pots with a spigot at the end for removing the excess water were prepared for vermicompost production. At first, some gravel and sand were added to pots as a drain. Two kilograms of cow manure were added to each pot. Some pots were enriched with phosphate rock (1%) or fishmeal (1%) or both of them. Each pot inoculated with 10 healthy adult earthworms of *Eiseniafoetida* with clear clitellum. After 2 weeks, bacterial treatment was inoculated with 5 mL of PSB inoculum ($1\times10^7 \frac{CFU}{ml}$). The pots were watered up to 60% of water holding capacity with distilled water to keep the pot moist. After 3 months, the whole materials were converted into a dark and fine texture product. Number and weight of earthworms and also number of cocoons were measured and collected. Samples of vermicompost were air-dried, passed through a 2-mm sieve, mixed uniformly and used for future analysis.

Table 1. Some chemical properties of cow manure and fishmeal

	N (%)	P (mg/kg)	Fe (mg/kg)	Mn (mg/kg)	Zn (mg/kg)	Cu (mg/kg)	pH	EC(dS/m)
Cow manure	2.59	8231.25	4372.875	309.6	161.75	27.5	8.26	2.34
Fishmeal	10.6	16875	3445.5	14.1	88.25	11.3	8.7	2.3

Table 2. Some chemical properties of phosphate rock

Cd (mg/kg)	Mn (mg/kg)	Fe (mg/kg)	Pb (mg/kg)	Cu (mg/kg)	Zn (mg/kg)	N (%)	P_2O_5 (%)
0.047	1.12	3.63	1.35	0.03	0.1	0.033	37

Total N was estimated by Micro-Kjeldahl method (Bremner, 1996) and total phosphorus by the Vanadate-Molybdate yellow method. Phosphorous was determined as PO_4^{3-} by the vanadium phosphomolybdate (vanadate colorimetry method) in which the phosphorous present as the orthophosphate reacts with a vanadate molybdate reagent to produce a yellow – orange complex, the absorbance of which was measured at 420nm (Chapman and Pratt, 1961). Micro-nutrients such as Fe, Cu, Mn, and Zn were determined using the dry ash method, dissolving in 2N HCl and then measuring with atomic absorption Shimadzu-AA670 (Chapman and Pratt, 1961). pH (Thomas, 1996) and EC (Rhoades, 1996) were measured in the ratio of 5:1 of water: vermicompost suspensions. The data were analyzed using SAS 9.1 statistical program by 3 ways ANOVA. Means were compared by least significant difference (LSD) at 5% level of significance using SAS statistical software. Graphs were prepared using Excel software.

RESULTS AND DISCUSSION

Total Nitrogen (N) Concentration

The effect of phosphate rock, fishmeal, and all interactions were significant on total N concentration in vermicompost (Table3). Phosphate rock decreased total N concentration by 12%, while application of fishmeal increased total N concentration by 4% in comparison with control (Figs.1 and 2).

Concentration was obtained in bedding material inoculated with the bacterium and enriched with fishmeal in the absence of phosphate rock (Table 4). Co-application of phosphate rock and fishmeal in non-bacterial and bacterial treatments significantly decreased total N concentration compared to non-phosphate rock treatments. The highest total N concentration was obtained in combined application treatments of PSB and fishmeal in non-phosphate rock treatments (Table 4).

Total Phosphorus (P) Concentration

The effect of phosphate rock, PSB, fishmeal and also interactions of phosphate rock and PSB (PR×B) and PSB and fishmeal (B×FM) were significant on total P concentration in vermicompost (Table 3). Adding all treatments to bedding materials caused a significant increase in P concentration. Application of PSB, phosphate rock, and fishmeal significantly increased P concentration by 4, 7 and 6%, respectively compared to the control ones (Table 4, Figs.1 and 2). Co-application of phosphate rock and PSB inoculation significantly increased total P concentration by 12% compared to the control (Table 4). Co-application of PSB and fishmeal significantly increased total P concentration in comparison with individual application of fishmeal or PSB.

Co-application of PSB and phosphate rock significantly increased total P concentration in comparison with individual application of PSB or phosphate rock. The maximum P concentration was obtained in the treatment of phosphate rock, fishmeal and PSB, which increased total P concentration by 20% compared to the control. Simultaneous application of phosphate rock, fishmeal, and PSB significantly increased P concentration in comparison with co-application of phosphate rock with fishmeal or PSB (Table 4).

pH

Analysis of variance (ANOVA) showed that the effect of phosphate rock and also interactions of PSB and fishmeal (B×FM), phosphate rock and fishmeal (PR×FM) and phosphate rock, PSB, and fishmeal (PR×B×FM) were significant on pH (Table 3). Application of phosphate rock and fishmeal significantly increased pH by 2% and 4%, respectively compared to the control (Figs. 1 and 2). Application of PSB along with fishmeal in non-phosphate rock treatments significantly decreased pH compared to the single application of fishmeal (Table 4). Simultaneous application of phosphate rock and fishmeal significantly decreased pH in non-bacterial treatments and significantly increased pH in bacterial treatments compared to the single application of fishmeal. The highest pH was obtained in the single application of fishmeal which increased by 11% compared to the control and the minimum pH was in the control.

Table 3. Analysis of variance of the effect of phosphate rock (PR), fishmeal (FM) and PSB (B) on some chemical properties of vermicompost.

SOV	df	Mean Square							
		N	P	pH	EC	Mn	Fe	Zn	Cu
PR	1	0.35**	570416**	0.25**	19.5**	46662.2**	32358^{ns}	70389**	988.8^{ns}
B	1	0.002^{ns}	326666*	0.01^{ns}	0.35^{ns}	1807^{ns}	504382^{ns}	158.8^{ns}	1495.4^{ns}
FM	1	0.02*	426666**	0.01^{ns}	2.40^{ns}	40413**	3839400**	16341**	809.1^{ns}
PR×B	1	0.02*	30104*	0.0009^{ns}	0.88^{ns}	694.9^{ns}	1803057*	648.40^{ns}	16540*
PR×FM	1	0.11**	12604^{ns}	0.15*	0.26^{ns}	49736**	572190^{ns}	14763**	18701*
B×FM	1	0.05**	65104**	0.44**	6.85*	7973.4^{ns}	902391^{ns}	14393**	11746^{ns}
PR×B×FM	1	0.03*	416.60^{ns}	0.71**	0.41^{ns}	6335.8^{ns}	1794023*	6792.2**	3910.4^{ns}
Error	16	0.01	25000	0.02	0.91	2143.6	320222	701.9	3650.9

ns, *, ** indicate not significant at P ≤0.05, significant at P ≤0.05 and P ≤0.01, respectively.

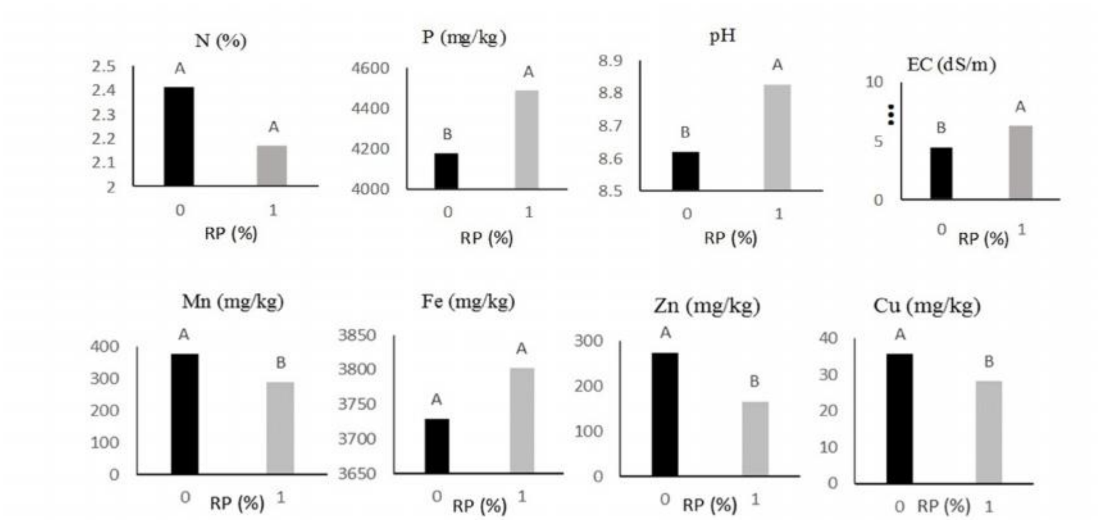

Fig. 1. Effect of phosphate rock (PR) on chemical properties of produced vermicompost.
Values followed by the same letter are not significantly different by LSD ($P \leq 0.05$).

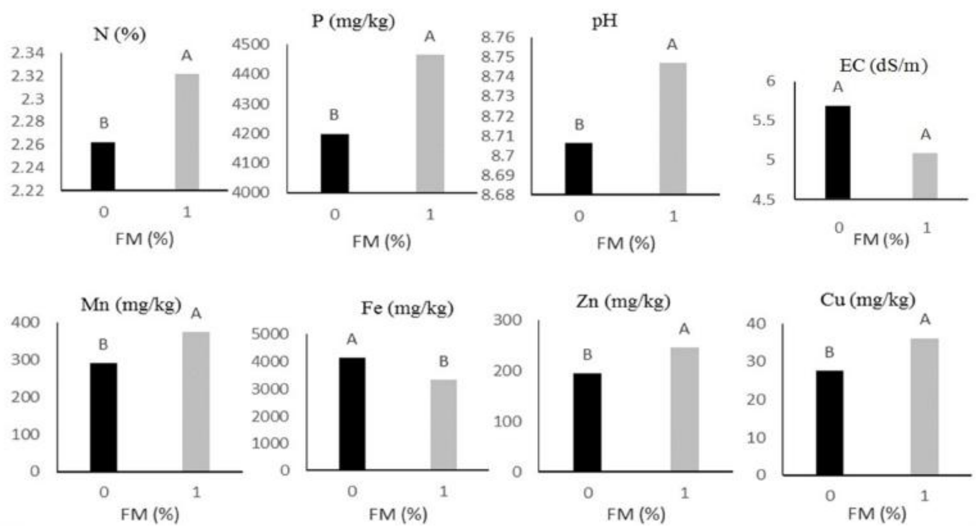

Fig. 2. Effect of fishmeal (FM) on chemical properties of produced vermicompost.
Values followed by the same letter are not significantly different by LSD ($P \leq 0.05$).

EC

The effect of phosphate rock and also the interaction of PSB and fishmeal (B×FM) were significant on EC (Table 3). Application of phosphate rock significantly increased EC by 40% in comparison with the control (Figure 1). Co-application of PSB and phosphate rock in non-fishmeal and fishmeal treatments significantly increased EC compared to the single application of PSB (Table 4). Simultaneous application of PSB and fishmeal in PR treated samples significantly decreased EC in comparison with single application of PSB. The maximum EC was obtained in the co-application of phosphate rock and PSB, which was higher than the control by 60%. The lowest EC could be seen in the combined application of PSB and fishmeal that was not significantly different from the control.

Total Mn Concentration

Total Mn concentration was significantly affected by phosphate rock (PR), fishmeal (FM) and the interaction of phosphate rock and fishmeal (PR×FM) (Table 3). Application of phosphate rock significantly reduced Mn concentration by 23% compared to the control but adding fishmeal increased Mn concentration by 27% compared to the control (Figs. 1 and 2). Inoculation of PSB did not have any significant effect on Mn concentration. Co-application of PR and fishmeal in non-bacterial and bacterial treated samples significantly decreased Mn concentration compared to the single application of fishmeal or co-application of fishmeal and PSB in non-phosphate rock treatments (Table 4). The maximum Mn concentration was obtained in the co-application of fishmeal and PSB ($R_0B_1F_1$) which was increased by 42% in comparison with the control (Table 4).

Table 4. Effect of phosphate rock, fishmeal and PSB on some chemical properties of vermicompost.

	treatment				
	PR_0		PR_1		Mean
	PR_0	PR_1	PR_0	PR_1	
	FM_0	FM_1	FM_0	FM_1	
B_0	2.43 b	2.47 ab	2.19 cd	2.10 d	2.30 A
B_1	2.19 cd	2.55 a	2.22 c	2.16 cd	2.28 A
			P (mg/kg)		
B_0	4000 e	4200 d	4275 cd	4400 bc	4258 B
B_1	4050 e	4475 b	4483 b	4800 a	4441 A
			pH		
B_0	8.1 d	9.0 a	8.9 ab	8.7 bc	8.70 A
B_1	8.8 ab	8.4 cd	8.8 ab	8.8 ab	8.75 A
			EC (dS/m)		
B_0	4.6 cd	5.0 bcd	6.0 abc	6.4 ab	5.5 A
B_1	4.8 cd	3.5 d	7.4 a	5.3 bc	5.3 A
			Mn (mg/kg)		
B_0	341.17 b	445.33 a	300.77 bc	287.83 bc	343.77 A
B_1	244.10 c	486.17 a	290.22 bc	285.20 bc	326.42 A
			Fe (mg/kg)		
B_0	3089.3 c	3532.8 bc	4566.5 a	3298.8 bc	3621.8 A
B_1	4862.0 a	3436.3 bc	4149.3 ab	3199.5 bc	3911.8 A
			Zn (mg/kg)		
B_0	274.92 b	294.08 b	172.17 bc	159.42 c	225.15 A
B_1	176.75 bc	361.17 a	162.08 bc	180.00 bc	220.00 A
			Cu (mg/kg)		
B_0	35.53 bc	47.15 a	26.16 cd	28.50 c	34.33 A
B_1	17.53 d	43.03 ab	32.10 c	27.01 cd	29.92 A

*Values followed by the same capital or small letter are not significantly different at LSD test ($P \leq 0.05$)

B_0: non-bacterial inoculation, B_1: inoculation with *Pseudomonas fluorescens*, F_0: without fishmeal, F_1: 1% fishmeal, R_0: without phosphate rock, R_1: 1% phosphate rock.

Total Fe Concentration

Analysis of variance (ANOVA) showed that the effect of fishmeal and also the interaction of phosphate rock and PSB (PR×B) and fishmeal, phosphate rock and PSB (PR×B×FM) were significant on total Fe concentration (Table 3). Fe concentration significantly increased by 57% in the individual application of bacterial treatments ($B_1F_0R_0$ vs. $B_0R_0F_0$) (Table 4). Adding phosphate rock also increased total Fe concentration by 47% compared to the control (Fig. 1). Fishmeal application significantly reduced Fe concentration by 19 % (Fig. 2). Combined application of PSB and fishmeal significantly decreased Fe concentration compared to the single application of PSB or phosphate rock (Table 4). Co-application of PR and fishmeal in bacterial and non-bacterial treatments significantly decreased Fe concentration compared to single PR treated samples (Table 4). The maximum Fe concentration was obtained in inoculation of PSB ($R_0B_1F_0$) that was not significantly different from the application of phosphate rock ($R_1B_0F_0$) and co-application treatments of phosphate rock and PSB($R_1B_1F_0$) (Table 4).

Total Zn Concentration

The effect of phosphate rock (PR) and fishmeal (FM) and interaction of phosphate rock and fishmeal (PR×FM), PSB and fishmeal (PSB×FM) and phosphate rock, PSB and fishmeal (PR×B×FM) were significant on total Zn concentration (Table 3). Co-application of fishmeal and PSB significantly increased Zn concentration by 31% compared to the control or individually PSB or fishmeal treated samples in non-phosphate rock treatments (Table 4). Zn concentration significantly decreased in the co-application of phosphate rock and fishmeal by 34% in comparison with the control or individually applied fishmeal (Table 4). Adding phosphate rock decreased Zn concentration by 64% (Fig. 1) while fishmeal application increased Zn concentration by 26% compared to the control (Fig. 2). The maximum Zn concentration was obtained in the co-application of PSB and fishmeal which was increased by 31% compared to the control.

Total Cu Concentration

Interactions of phosphate rock and PSB (PR×B) and phosphate rock and fishmeal (PR×FM) had a significant effect on total Cu concentration (Table 3). Adding phosphate rock decreased Cu concentration by 20% while adding fishmeal increased it by 30% compared to the control (Figs. 1 and 2). Co-application of PR and PSB significantly increased Cu concentration in non-fishmeal treatments compared to single PSB inoculated treatments (Table 4). However, simultaneous application of PR and PSB in fishmeal treatments significantly decreased Cu concentration. In non-bacterial and bacterial treatments, combined application of PR and fishmeal significantly decreased Cu concentration in comparison with single fishmeal treated samples. The maximum concentration of Cu was obtained at the treatment of fishmeal which was 32% higher than the control.

Number of Adult Earthworms

The result of ANOVA (Table 5) showed that effects of all treatments and their interactions on the number of adult earthworms were significant. Bacterial inoculation and the application of PR significantly decreased the number of earthworms by 20% and 43% compared to control treatments (Table 6 and Fig. 3). Application of fishmeal significantly increased the number of earthworms by 11% compared to the control (Fig. 4).

Co-application of PR and fishmeal in non-bacterial and bacterial treated samples significantly decreased the number of adult earthworms compared to the individual application of fishmeal or PSB (Table 6). Simultaneous application of PSB and fishmeal in both non-PR and PR treatments significantly decreased the number of adult earthworms compared to the individual application of PSB and the single application of fishmeal. In non-bacterial and bacterial treatments, combined application of PR and fishmeal significantly increased the number of adult earthworms compared to the individual application of PR. The highest number of adult earthworms was obtained at the treatments of single application of fishmeal and the minimum number of adult earthworms was obtained at the co-application treatment of PR and PSB.

Table 5. Analysis of variance of the effect of phosphate rock (PR), fishmeal (FM) and PSB (B) on the number and weight of earthworms and number of cocoons

		Mean Square		
SOV	df	Number of earthworms	Number of cocoons	Earthworms weight
PR	1	7107.04**	513.37**	1863.48**
B	1	1218.37**	63.37**	317.04**
FM	1	273.37**	22.04**	70.52**
PR×B	1	210.04**	5.04**	54.74**
PR×FM	1	477.04**	26.04**	125.19**
B×FM	1	30.37*	9.37*	7.82*
PR×B×FM	1	30.37*	2.042*	7.68*
Error	16	4.96	0.92	1.28

ns, *, ** indicate not significant at P ≤0.05, significant at P ≤0.05 and P ≤0.01, respectively.

Table 6. Effect of PR, fishmeal, and PSB on the number and weight of earthworms and number of cocoons

Treatment					
	PR_0		PR_1		
	FM_0	FM_1	FM_0	FM_1	Mean
			Number of earthworms		
B_0	87.67 a	90 a	40.67 f	56.333 d	68.67A
B_1	72b	65.333 c	32.33 g	48 e	54.42 B
			Number of cocoons (in 20 g of vermicompost)		
B_0	20.33 b	22 a	8.67 g	13.33 e	16.08 A
B_1	18 c	16 d	7 b	10.33 f	12.83 B
			Earthworms biomass (earthworms weight)		
B_0	44.87 a	46 a	20.79 f	28.79 d	35.11 A
B_1	36.85 b	33.44 c	16.55 de	24.54 e	27.84 B

*Values followed by the same capital or small letter are not significantly different at LSD test (P ≤0.05)

B_0: non-bacterial inoculation, B_1: inoculation with *Pseudomonas fluorescens*, F_0: without fishmeal, F_1: 1% fishmeal, R_0: without phosphate rock, R_1: 1% phosphate rock.

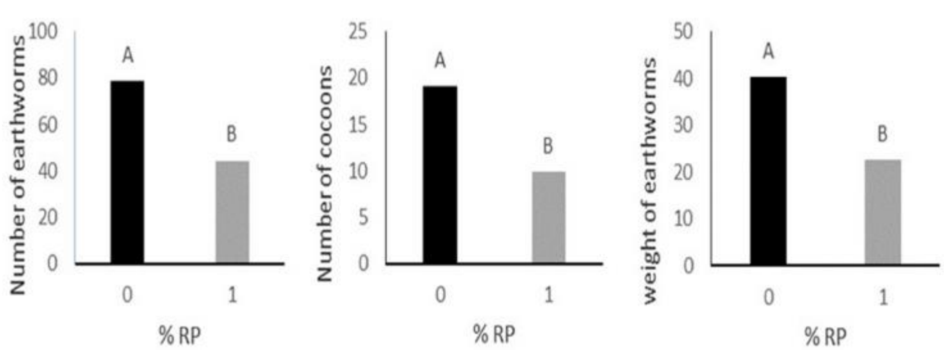

Fig. 3. Effect of phosphate rock (PR) on the number and weight of earthworms and number of cocoons. Values followed by the same letter are not significantly different by LSD (P ≤0.05).

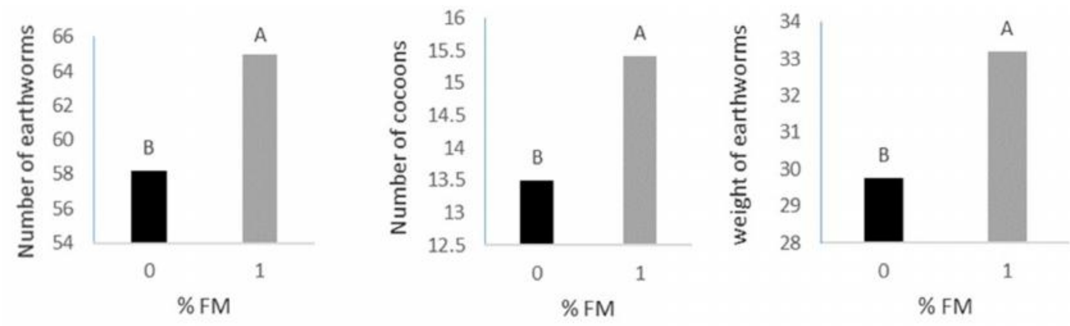

Fig. 4. Effect of fishmeal (FM) on the number and weight of earthworms and number of cocoons. Values followed by the same letter are not significantly different by LSD ($P \leq 0.05$).

Number of Cocoons

Effect of PR (PR) and PSB inoculation (B) and also the interactions of PR and PSB (PR×B), PSB and fishmeal (B×FM) and PR, PSB and fishmeal (PR×B×FM) were significant on the number of cocoons (Table 5). Application of PSB and PR significantly decreased the number of cocoons by 20% and 43%, respectively (Table 6, Figure 3). Fishmeal application increased the number of cocoons by 14% compared to the control (Fig. 4).Co-application of PR and fishmeal in non-bacterial and bacterial treated samples significantly decreased the number of cocoons compared to the single application of fishmeal (Table 6). Combined application of PR and PSB significantly increased the number of cocoons compared to the single application of PSB. In bacterial treatments, simultaneous application of PR and fishmeal significantly decreased the number of cocoons compared to the single application of PR. Co-application of PR and PSB in non-fishmeal treatments significantly increased the number of cocoons compared to the single application of PR. The maximum number of cocoons was obtained in individually fishmeal treated samples.

Weight of Adult Earthworms

Results of Table 5 showed that effects of all treatments and their interactions on the weight of adult earthworms were significant. Application of PR and also PSB inoculation significantly decreased the weight of adult earthworms by 43% and 21%, respectively compared to control ones (Figure 3 and Table 6). Application of fishmeal significantly increased the weight of adult earthworms by 11% compared to the control (Figure 4). Co-application of PR and fishmeal in non-bacterial and bacterial treated samples significantly decreased the weight of adult earthworms compared to the single application of fishmeal and increased in comparison with individually PR treated samples (Table 6). Simultaneous application of PSB and fishmeal in non-PR treatments significantly decreased the weight of adult earthworms compared to the individually applied PSB or fishmeal. Co-application of PR and PSB in non-fishmeal and fishmeal treatments significantly decreased the weight of earthworms compared to the single application of PSB or PR. The maximum weight of adult earthworms was obtained at the control treatment.

Results showed that adding phosphate rock and PSB inoculation reduced nitrogen concentration that was consistent with previous studies. Chaudhuri et al. (2000), for example, observed that total nitrogen reduced during vermicompost production. They reported that this reduction was because of ammonia volatilization, its adsorption to earthworms and leaching. Increasing pH due to the application of phosphate rock may decrease N concentration. Nitrogen could volatilize in alkaline pH as ammonia (Hartenstein, 1981).

Enriching bedding materials by phosphate rock, fishmeal, and PSB, for vermicompost production increased total phosphorus. Increase in total phosphorus in vermicompost was higher when phosphate rock was used with PSB inoculation. PSB activities in vermicompost can help phosphorus solubilizing due to releasing organic acids and chelating agents (Kumar and Narula, 1999). Kumar and Singh (2001) reported that inoculation of *Pseudomonas striata* to vermicompost caused an increase in available phosphorus in vermicompost. El-Haddad et al. (2014) reported that adding phosphate rock to rice straw caused an increase in total phosphorus in vermicompost and vermicompost tea. Adding fishmeal with PSB inoculation increased total phosphorus concentration in vermicompost, which may be due to phosphatase activities of used bacteria. Organic phosphorus mineralizes with phosphatase enzymes which are released by PSBs (Eivazi and Tabatabai, 1977). Earthworms can help microbial growth and enzyme activities and promote phosphorus solubilization (Wana and Wang, 2004).

Overall, the pH was between 8 and 9. Edwards (1995) reported the best pH for vermicompost production as between 5 and 9. Microorganisms and earthworm may mineralize nitrogen-containing organic matters during vermicomposting and produce alkaline compounds like ammonia that could increase pH (Kharazi et al. 2012). Phosphate rock caused increasing in EC of vermicompost. That was consistent with previous studies. Increasing EC in vermiculture technology is due to releasing available ions through digesting bedding material by earthworms (Mirbolook et al., 2011). During composting and vermicomposting, some exchangeable minerals such as Ca, Mg and K release in available forms and so increase EC (Guoxue et al. 2001; Tognetti et al. 2005). Pattnaik and Reddy (2010) reported increased EC during the period of vermicomposting green waste.

Sharma et al. (2005) reported that the concentration of metals such as Zn and Cu decreased through vermiculture technology by adsorbing to earthworms body and leaching. On the other hand, due to decomposition of bedding material, the concentration of these metals may increase in vermicompost (Deolalikar et al., 2005). Also, researches showed that decomposer bacteria caused an increase in metals concentration in vermicompost (Saha et al., 2008). Dominguez et al. (1997) reported that although mineralization of carbon increased metals during vermicomposting, the available forms decreased. Vermicompost producing from sewage sludge increased Fe concentration, but Cu, Mn, and Zn concentrations decreased because of accumulation in earthworm body (Khwairakpam and Bhargava, 2009). PSB may increase Fe concentration due to the production of siderophore.

Results showed that the PR and PSB inoculation had negative influences on the number and weight of earthworms and also the number of cocoons. The number of cocoons shows earthworm's tendency to reproduction (Mirbolook et al., 2008). The differences between the measured parameters in treatments depend on the biochemical quality of the earthworm's growth context (Flack and Hartenstein, 1984). The reduction in the number of earthworms may be due to the presence of Cd and Pb in used PR, which leads to a reduction in weight and their reproduction of earthworms. Edwards et al. (1988) reported that these stresses have negative influences on earthworm's growth and reproduction. Hartenstein et al. (1981) found that in toxic conditions, *E. fetida* can move constantly into areas free of its castings. Also, the significant decrease in the number of earthworms and cocoons and weight of earthworms may be due to the lack of oxygen because of the high activities of these bacteria. Earthworm's respiration is by their skin, and the lack of oxygen can naturally affect their biomass and reproduction (Mirbolook et al., 2008; Mendes and Almedia, 1962).

CONCLUSION

Results showed that adding phosphate rock to bedding materials of vermicompost increased P and Fe concentrations. Inoculation of PSB increased total P with solubilizing organic and inorganic phosphorus and Fe concentration with siderophore production. Fishmeal was more effective on enriching vermicompost, due to increasing N, P, Mn, Cu and Zn concentrations in vermicompost and the effect of fishmeal was more when it was used with PSB inoculation. Also, results showed that application of PR and PSB decreased the number of earthworms and cocoons and the weight of earthworms. Application of fishmeal increased the measured parameters. Overall, it should be concluded that application of all treatments had positive effects on chemical properties of vermicompost and was favorable for the vermicomposting process. But, application of PR and PSB negatively affected the earthworms and their productivity; therefore, they were not suitable for vermiculture process.

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دانشگاه شیراز

تاثیر افزودنی‌های آلی و معدنی بر برخی خصوصیات شیمیایی ورمی‌کمپوست، زیست توده و تولید مثل کرم‌های خاکی

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کوکون

پودر ماهی

باکتری حل کننده فسفات

خاک فسفات

چکیده-غنی سازی بستر در سیستم تولیدی ورمی کمپوست با افزودنی های آلی و معدنی می تواند کیفیت ورمی کمپوست تولیدی را افزایش دهد. خاک فسفات و پودر ماهی مواد ارزان و قابل دسترس هستند و مایه زنی بستر با ریزجانداران حل کننده فسفات می تواند قابلیت استفاده عناصر غذایی آنها را افزایش دهد. به منظور بررسی تاثیر غنی سازی بستر تولید ورمی کمپوست با خاک فسفات، پودر ماهی و باکتری حل کننده فسفات، آزمایش فاکتوریل در غالب طرح کاملا تصادفی با سه تکرار انجام گردید. عوامل آزمایش شامل دو سطح باکتری حل کننده فسفات (*سودوموناس فلورسنس*) (بدون مایه زنی و مایه زنی شده)، دو سطح خاک فسفات (صفر و ۱ ٪ وزنی) و دو سطح پودر ماهی (صفر و ۱٪ وزنی) بود. نتایج نشان داد که کاربرد خاک فسفات و باکتری موجب کاهش غلظت نیتروژن گردید در حالی که کاربرد پودر ماهی غلظت نیتروژن را در ورمی‌کمپوست افزایش داد. کاربرد خاک فسفات، پودر ماهی و باکتری به تنهایی و همزمان سبب افزایش غلظت فسفر شد. کاربرد پودر ماهی موجب افزایش غلظت مس، روی و منگنز شد. خاک فسفات موجب افزایش غلظت آهن و همچنین کاهش مس، روی و منگنز شد. کاربرد خاک فسفات موجب افزایش قابلیت هدایت الکتریکی و کاربرد پودر ماهی موجب کاهش آن شد. به علاوه خاک فسفات و پودر ماهی هردو موجب افزایش پ هاش شدند. از طرف دیگر کاربرد خاک فسفات و باکتری حل کننده فسفات موجب کاهش تعداد و وزن کرم‌های خاکی بالغ و همچنین تعداد کوکون گردید. کاربرد پودر ماهی تعداد و وزن کرم و تعداد کوکون را افزایش داد. نتایج نشان داده است که افزودن مواد به بستر قابلیت دسترسی برخی عناصر غذایی را افزایش و رشد و زیست توده کرمهای خاکی را تغییر داده است.