Journal of Environment and Earth Science
ISSN 2224-3216 (Paper) ISSN 2225-0948 (Online)
Vol. 3, No.12, 2013

Effect of Different Bulking Materials and Earthworms Species on Bioremediation Potential of Municipal Sewage Sludge

Hossein Azarpira[1*], Pejman Behdarvand[2] & Kondiram Dhumal[1]
1. Department of Environmental Sciences, University of Pune, Pune-07-India
2. Department of Agricultural and Natural Resources, Ahvaz Branch, Islamic Azad University, Ahvaz, Iran
* E-mail of the corresponding author: hazarpira912@gmail.com

Abstract
In present study the efficiency for vermiremediation of municipal sewage sludge (MSS) with different bulking materials such as Cow Dung (CD), Sheep Manure (SM) and Garden Soil (GS) in presence of *Eudrilus eugeniae* and *Eisenia foetida* was investigated. The results revealed that interaction of worm species and bulking materials had significant effect on moisture %, EC and Cl^- but it was non significant for pH and Organic Carbon (OC). At final stage of vermiremediation OC, EC and Cl^- were reduced as compare to initial stage. The highest values for EC (8804 µmohs/cm) and OC (25.63 %), were noted in the treatment SS + CD and no worm species. While maximum Cl^- (1423 ppm) was found in treatment SS + no bulking material and no worm species. The results also indicated that the lowest EC (3424 µmohs/cm) was in treatment SS + GS + *Eudrilus eugeniae*, OC (13.83 %) was in treatment SS + no bulking material + *Eudrilus Eugeniae* and Cl^- (643.0 ppm) was noted in treatment SS + CD + *Eudrilus eugeniae.* It is concluded that the effective bioremediation of sewage sludge is possible with cow dung manure or garden soil along with *Eudrilus eugeniae* or *Eisenia foetida*.
Keywords: bulking, earthworm, remediation, sewage sludge

1. Introduction

The wastewater treatment is concentrated on the treatment of sewage water rather than sewage sludge. Sewage sludge is always dumped on the open grounds and left alone for natural degradation. Most of the times sewage sludge is directly applied to the crop land as fertilizer. Different studies (Smith *et al.* 2001; Oleszczuk 2006) indicated that its application can be harmful to soil and plants if directly used. Heavy metals and organic compounds are the most serious pollutants present in the sewage sludge Stevens *et al.* (2003). Most of the sewage treatment plants have only primary wastewater treatment facilities (sedimentation), which generate high quantities of primary sewage sludge. Such huge quantities of sewage sludge appear to be serious environmental pollution issue in developing countries. Sludge production is unavoidable problem arising from the treatment of municipal wastewater. This sludge contains considerable amounts of organic matter, pathogens and chemical contaminates. If it is not properly managed may create extensive health hazards Mesdaghinia *et al.* (2004). Amongst the several available alternatives for disposal of sewage sludge, one of the most convenient is using it in agriculture. Sewage may be a good plant growth substance, if undesired chemicals are removed from sludge through vermiremediation system. In recent years, the potential of earthworms has been tested to stabilize the sewage sludge from urban and industrial localities Suthar (2010).

Vermiremediation is most accepted and highly beneficial method for the management of sewage sludge. Vermicomposting involves the defragmentation and partial digestion of organic waste by earthworms, and further digestion by exogenous hydrolytic enzymes provided by earthworm gut-associated microflora. Recent studies indicated the potential of earthworms in stabilization of urban sewage sludge. In the vermicomposting process, earthworms maintain aerobic condition in the organic wastes and convert a portion of it into biological biomass and respiration products, and expel the remaining stabilized product, i.e. vermicompost Khwairakpam & Bhargava (2009). Therefore the present study was conducted to investigate the effects of different bulking materials such as cow dung, sheep manure and garden soil on the remediation potential of two earthworm species viz *Eudrilus eugeniae* and *Eisenia foetida*.

2. Materials and Methods

2.1 Collection of samples

The sewage sludge samples were collected from sewage treatment plant situated in Pune city (Bopodi area) during the year 2012 in the month of Jan-Feb and brought to the laboratory for the remediation study. After sun drying these samples were crushed into fine powder which was passed through 0.5 mm size sieve Gupta (2007). These samples were analyzed for pH, EC, Moisture, OC and Cl^-. The bulking materials such as cow dung, sheep manure and garden soil were collected from local sources. The species of earthworm viz *Eisenia foetida* and *Eudrilus eugeniae* were obtained from *Vasantdada Sugar Institute (*VSI) and Institute of Natural Organic Agriculture (INORA), Pune.

2.2 Experimental Design

The vermiremediation experiment was conducted at Department of Environmental Science, University of Pune, India, by using factorial arrangement with randomized complete block design with three replications. Treatments included W0: no worm, W1: *Eisenia foetida* and W2: *Eudrilus eugeniae*; and for bulking materials (B0: sewage sludge, B1: sewage sludge + cow dung, B2: sewage sludge + sheep manure and B3: sewage sludge + garden soil). The earthen pots (25x25x30cm) were filled with sewage sludge and bulking materials in proportion of 2:1 along with 50 earthworms of species *Eisenia foetida* and *Eudrilus eugeniae*. The experiment was carried out for 90 days.

2.3 Physico-chemical Analysis

Moisture % in sludge and vermicompost was determined using 25 g of sample from each treatment, by drying it at 103-105°C for 18 to 23 h in hot air oven till the constant weight was recorded. OC was measured after ignition of the sample in muffle furnace at 550°C for 1 h Nelson & Sommers (1996). The pH and EC were measured in 1:10 (w/v) suspension of samples in de-ionized water which was mixed at 230 rpm for 30 minutes and filtered through Whatsman No. 1 filter paper APHA AWWA WEF (1995). For analysis of Cl^- 25 g dried sample was taken and to it 50 ml distilled water was added. After shaking and filtering the sample, the filtrate was used for further analysis Gupta (2007).

2.4 Statistical Analysis

The recorded data was analyzed statistically by using MSTATC computer software and a comparison of recorded data was done on the basis of Duncan's multiple range tests at Alfa level 5%.

3. Results and Discussion

3.1 Organic Carbon

The analysis of data showed that interaction of worm species and bulking materials had non significant effect on organic carbon (OC) at final stage of vermiremediation (Table 1). As compare to initial stage OC was decreased in final stage. The highest OC (25.63 %) was recorded in SS + CD in absence of worm species in final stage (Table 2). The increase of OC in absence of earthworms also reported by Ndegwa & Thomson (2000). The highest carbon content may be due to highest carbon in cow dung. As there was no worm species there was no consumption of carbon and no release of CO_2 through their respiration. Similarly there was no digestion of carbohydrates and polysaccharides from the substrates (cow dung) by earthworms Suthar (2010). The increase in OC % in the present investigation may be attributed to above mentioned reasons.

The lowest OC (13.83 %) was noted in treatment SS + no bulking material and in presence of *E. eugeniae* in final stage (Table 2). Similar results were reported by Yadav & Garg (2011) during vermicomposting of organic wastes. The combined action of earthworms and microorganisms may be responsible for loss of OC in the form of CO_2 Prakash & Karmegam (2010). Similar may be the reason for decrease in OC during vermicomposting of sewage sludge. Azizi *et al.* (2013) also recorded reduction in OC during vermicomposting of sludge. They further explained that loss in OC was due to use of OC by earthworms and microorganisms as source of energy, organic matter stabilisation in the substrate as a result of combined action of earthworms and microorganisms.

3.2 Moisture

The statistical analysis of data revealed that interaction of worm species and bulking materials had significant effect on moisture % at final stage of vermiremediation (Table 1). The moisture % was higher at final stage of vermicomposting as compare to initial stage. At final stage of vermicomposting, the maximum moisture (60.83 %) was reported in SS + CD in presence of *Eudrilus eugeniae* (Table 2).

Patnaik & Reddy (2010) also reported similar trend for moisture % in presence of different species of earthworms during vermicomposting. According to Yadav & Garg (2011) adequate moisture content is one of the most important factors necessary for the biological activities of earthworms and microorganisms in vermicomposting process. It also acts as a medium for different chemical reactions and transport of nutrients. The ideal moisture % range is 60-80 % (Edwards 1998). In present study the highest moisture % recorded was in the above range (60.83 %), which may be helping the worm species for various biological activities.

The minimum moisture % (43 %) was noted in treatment SS + SM in absence of worm species (Table 2). Similar finding was reported during vermicomposting of sugar mill and distillery effluents Rai & Singh (2012).

Table 1. Mean squares of variance analysis for organic carbon and moisture content of sewage sludge + bulking materials

Sources	df	OC content (%)		Moisture content (%)	
		Initial	Final	Initial	Final
Replication	2	9.810	2.59	6.44	2.92
Worm (W)	2	0.000 ns	6.87*	0.000 ns	128.552 **
Bulking (B)	3	213.84 **	176.23 **	405.593 **	279.017**
W × B	6	0.000 ns	2.706ns	0.000 ns	8.474*
Error	22	4.23	1.94	2.94	6.794
C. V (%)		8.23	7.46	3.70	5.16

*, **: significant at 5 and 1 %, respectively, ns: not significant

Table 2. Interaction effect of worm species and bulking materials on organic carbon and moisture % of vermiremediation

Treatment	OC content (%)		Moisture content (%)	
	Initial	Final	Initial	Final
W0B0	20.07 b	15.30 c	45.60 b	45.50 f
W0B1	30.37 a	25.63 a	56.03 a	55.10 bc
W0B2	21.73 b	15.70 c	42.80 bc	43.00 f
W0B3	27.73 a	21.53 b	41.03 c	43.27 f
W1B0	20.07 b	14.47 c	45.60 b	51.73 cde
W1B1	30.37 a	22.07 b	56.03 a	59.50 ab
W1B2	21.73 b	15.13 c	42.80 bc	47.60 ef
W1B3	27.73 a	21.77 b	41.03 c	52.10 cde
W2B0	20.07 b	13.83 c	45.60 b	52.57 cd
W2B1	30.37 a	22.03 b	56.03 a	60.83 a
W2B2	21.73 b	14.93 c	42.80 bc	47.87 def
W2B3	27.73 a	21.73 b	41.03 c	46.57 f

Means with different letters are significantly different at P=0.05, using Duncan's Multiple Range Test.

3.3 Chlorides

The results revealed that interaction of worm species and bulking materials had significant effect on chloride contents during vermicomposting (Table 3). Chloride content varied with interaction of worm species and bulking materials. The Cl^- content was reduced in final stage as compare to initial stage. Amongst the treatments, maximum Cl^- (1423 ppm) was noted in absence of worm species and bulking material at final stage (Table 4). The result of present investigation is in agreement with Shirazi & Marandi (2012) who reported that the chloride content in absence of worm species was very high.

The minimum chloride contents (643 ppm) were reported in SS + CD in presence of *Eudrilus eugeniae*. It was statistically followed by the treatment SS + CD in presence of *Eisenia foetida* (747.7 ppm) (Table 4). Similar results were reported by Reddy *et al* (2012) for chloride content of sewage sludge due to the use of different bulking materials. The earthworm species are capable to change the biochemical constituents like chlorides from higher to lower level. This may be due to enzymatic digestion, transformation of chemical constituents and microbial action on different organic and inorganic compounds in the sewage sludge Yadav & Garg (2011).

3.4 pH

The results shown in Table 3 indicated that worm species and bulking materials had no significant effect on pH during vermiremediation. The pH value at final stage was increased for all the treatments as compare to initial stage (Table 4). The pH greatly influences vermicomposting process and the acceptable pH range for biological activities of earthworms is (5.5–8.5). The evolution of CO_2 and utilization of volatile fatty acids, are responsible to increase the pH Yadav & Garg (2011). The increase in pH of vermicompost was in consistence with findings of Loh *et al.* (2005).

3.5 Electrical Conductivity

Worm species and bulking materials had significant effect on EC during vermicomposting process (Table 3). The EC values decreased during vermicomposting at final stage over initial stage (Table 4). The highest EC (8804 µmohs/cm) was in SS + CD and in absence of worm species. The findings of present study are in agreement with Lombard *et al.* (2011) who reported that increasing of EC in vermicomposting may be due to accumulation of soluble salts and release of different mineral salts in available forms such as phosphate, ammonium, potassium etc.

The lowest EC (3424 µmohs/cm) was noted in treatment SS + GS with *Eudrilus eugeniae* (Table 4). Singh *et al* (2010) also recorded decrease in EC during vermicomposting with worm species and bulking materials. The reduction in EC may be attributed to biochemical changes in sewage sludge by the activities of worm species.

Table 3. Mean squares of variance analysis for chloride, pH and electrical conductivity content of sludge + bulking material

Sources	df	Cl^- content (ppm)		pH		Ec (µmohs/cm)	
		Initial	Final	Initial	Final	Initial	Final
Replication	2	315.3	49.1	0.1	0.03	154563.3	145523.4
Worm (W)	2	0.000^{ns}	609370^{**}	0.000^{ns}	0.01^{ns}	0.000^{ns}	14999179^{**}
Bulking (B)	3	130566^{**}	196537^{**}	2.34^{**}	0.03^{*}	14792812^{**}	7381875^{**}
W × B	6	0.000^{ns}	29679^{**}	0.000^{ns}	0.02^{ns}	0.000^{ns}	1447520^{**}
Error	22	9438.9	5103.1	0.089	0.03	61011.9	139408.6
C. V (%)		7.17	6.68	4.48	2.44	3.24	6.29

*, **: significant at 5 and 1 %, respectively, ns: not significant

Table 4. Interaction effect of worm species and bulking materials on chloride, pH and electrical conductivity of vermiremediation

Treatment	Cl^- content (ppm)		pH		Ec (µmohs/cm)	
	Initial	Final	Initial	Final	Initial	Final
W0B0	1450.0 a	1423.0 a	6.3 b	6.9 a	7776.0 b	6941.0 bc
W0B1	1188.0 b	1159.0 b	6.5 b	7.1 a	9019.0 a	8804.0 a
W0B2	1346.0 ab	1321.0 a	6.4 b	7.0 a	7782.0 b	7361.0 b
W0B3	1435.0 a	1397.0 a	6.5 a	7.0 a	5910.0 c	5714.0 def
W1B0	1450.0 a	977.3 c	6.3 b	6.9 a	7776.0 b	5464.0 ef
W1B1	1188.0 b	747.7 de	6.5 b	7.1 a	9019.0 a	5849.0 de
W1B2	1346.0 ab	1044.0 bc	6.4 b	7.1 a	7782.0 b	5965.0 de
W1B3	1435.0 a	827.0 d	6.3 a	7.0 a	5910.0 c	4777.0 g
W2B0	1450.0 a	1057.0 bc	6.3 b	7.1 a	7776.0 b	5578.0 ef
W2B1	1188.0 b	643.0 e	6.5 b	7.1 a	9019.0 a	5043.0 fg
W2B2	1346.0 ab	1118.0 b	6.4 b	7.1 a	7782.0 b	6326.0 cd
W2B3	1435.0 a	1122.0 b	6.4 a	6.9 a	5910.0 c	3424.0 h

Means with different letters are significantly different at P=0.05, using Duncan's Multiple Range Test.

4. Conclusion

The serious issue of environmental pollution created by municipal sewage sludge may be resolved by using eco-friendly technology of vermiremediation. Use of bulking materials along with earthworms speed up the process, removing different environmental pollutants, heavy metals and chemical contaminants, leading to protection of the environment and safe use in agriculture. The results emerged from present investigation clearly indicated that cow dung along with *Eudrilus eugeniae* is the best combination for vermiremediation of municipal sewage sludge.

Acknowledgments

The authors are thankful to Head, Department of Environmental Sciences, University of Pune for providing research facilities and encouragement. We also thank the authorities of Pune Municipal Corporation for availing the samples of municipal wastewater sludge.

References

APHA, AWWA, WEF. (1995), "Standard Methods for the Examination of Water and Wastewater", 19th ed. American Public Health Association, American Water Works Association, Water Environment Federation, Washington DC, U.S.A.

Azizi, A.B., Lime, M.P.M., Noor, Z.M. & Abdullah, N. (2013), "Vermiremoval of Heavy Metal in Sewage Sludge by Utilising *Lumbricus Rubellus"*, *Ecotoxicology and Environmental Safety*, 90, 13-20. http://dx.doi.org/10.1016/j.ecoenv.2012.12.006.

Edwards, C.A. (1998), "The Use of Earthworms in the Breakdown and Management of Organic Wastes", In: Edwards CA (ed) Earthworm Ecology. St. Lucie Press, Boca Raton, pp 327–354.

Gupta, P.K. (2007), "Soil, Plant, Water and Fertilizer Analysis", 2nd Edition, Published by Agribios, Jodhpur. India 350 p.

Khwairakpam, M. & Bhargava, R. (2009), "Bioconversion of Filter Mud Using Vermicomposting Employing two Exotic and one Local Earthworm Species", *Bioresour. Technol*. 100, 5846–5852. http://dx.doi.org/10.1016/j.biortech.2009.06.038.

Loh, T. C., Lee, Y.C., Liang, J.B. & Tan, D. (2005), "Vermicomposting of Cattle and Goat Manures by *Eisenia Foetida* and their Growth and Reproduction Performance", *Bioresour. Technol* 96, 111-114. http://dx.doi.org/10.1016/j.biortech.2003.03.001.

Lombard, K., Neill, M., Heyduk, R., Onken. B., Ulery, A., Mexal, J. & Unc, A. (2011), "Composted Biosolids as a Source of Iron for Hybrid Poplars (*Populus sp*.) Grown in Northwest New Mexico", Agroforest Syst 81, 45-56. http://dx.doi.org/10.1007/s10457-010-9334-7.

Mesdaghinia, A.R., Panahi Akhavan, M., Vaezi, F., Naddafi, K. & Moosavi, G. (2004), "Waste Sludge Characteristics of a Wastewater Treatment Plant Compared with Environmental Standards", *Iranian J. Publ Health*, 33, (1) 5-9

Ndegwa, P.M. & Thomposon, S.A. (2000), "Effects of C to N Ratio on Vermicomposting of Biosolids", *J. of Bioresource Thchnology*. 75, 7-12. http://dx.doi.org/10.1016/S0960-8524(00)00038-9.

Nelson, D.W. & Sommers, L.E. (1996), "Total Carbon and Organic Carbon and Organic Matter", In: Page, A.L., Miller, R.H., Keeney, D.R., (Eds.) Method of Soil Analysis. *American Society of Agronomy*, Madison, pp. 539-579.

Oleszczuk, P. (2006), "Characterization of Polish Sewage Sludges with Respect to Fertility and Suitability for Land Application", *J. Environ. Sci. Health* A 41, 1119–1217.

Patnaik, S. & Reddy, M.V. (2010), "Nutrient Status of Vermicompost of Urban Green-Waste Processed by three Earthworm Species – *Eisenia Foetida*, *Eudrilus Eugeniae* and *Perionyx Excavates", Applied and Environmental Soil Science* Vol. (Article id 967526), 1-13. http://dx.doi.org/10.1155/2010/967526.

Prakash, M. & Karmegam, N. (2010),"Vermistabilization of Pressmud Using *Perionyx ceylanensis*mich",*BioresourTechnol*,10,8464–8468. http://dx.doi.org/10.1016/j.biortech.2010.06.002.

Rai, R. & Singh, K. (2012), "Physic-chemical Analysis and Management of Different Combinations of Sugar Mill Distillery Effluents with Different Animal Dung during Vermicomposting by Earthworm *Eisenia Foetida", Journal of Biology, Agriculture and Healthcare*. 2,(1).

Reddy, S. A., Akila, S. & Kale, R. D. (2012), "Management of Secondary Sewage Sludge by Vermicomposting for Use as Soil Amendment", *Global Journal of Biotechnology & Biochemistry* 7 ,(1), 13-18.

Shirazi,E.K., & Marandi, R. (2012), "Evaluation of Heavy Metals Leakage from Concretes Containing Municipal Wastewater Sludge", *Enviro and Pollution*; Vol. 1, No. 2. http://dx.doi.org/10.5539/ep.v1n2p176.

Singh, J., Kaur, A., Vig, A.P. & Rup, P.J. (2010), "Role of *Eisenia foetida* in Rapid Recycling of Nutrients from Biosludge of Beverage Industry". *Ecotoxicol Environ Saf*. 73(3), 430-5. http://dx.doi.org/10.1016/j.ecoenv.2009.08.019.

Smith, K.E., Green, M., Thomas, G.O. & Jones, K.C. (2001), "Behavior of Sewage Sludge-Derived PAHs on Pasture", *Environ. Sci. Technol*. 35, 2141–2150. http://dx.doi.org/10.1021/es000178l.

Stevens, J.L., Stern, G.A., Tomy, G.T. & Jones, K.C. (2003), "PAHs, PCBs, PCNs, Organochlorine Pesticides, Synthetic Musks and Polychlorinated N-alkanes in UK Sewage Sludge, Survey Results and Implications", *Environ. Sci. Technol.* 37, 462–467. http://dx.doi.org/10.1021/es020161y.

Suthar, S. (2010), "Recycling of Agro-industrial Sludge through Vermitechnology", *Ecological Engg* 36, 1028-1036. http://dx.doi.org/10.1016/j.ecoleng.2010.04.015

Yadav, A. & Garg, V.K. (2011), "Industrial Wastes and Sludges Management by Vermicomposting", *Rev Environ Sci Biotechnol* 10, 243-276.