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Pathogens' Reduction in Vermicompost Process Resulted from the Mixed Sludge Treatments-Household Wastes

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ABSTRACT

Introduction: The presence of pathogenic microbial agents and pathogens in organic fertilizers causes health problems and disease transmission. The aim of this study was to evaluate the efficiency of vermicomposting process in improve the microbial quality of the compost produced.
Materials and Methods: This experimental study was conducted as a pilot-scale one, in the laboratory of school of Health. In order to produce vermicompost, some perishable domestic waste were mixed whit sludge of wastewater treatment plant in a reactor. Tests to determine the microbial quality of the product were carried out at an early stage, during the process of production, and on the final product. The worms used for the production of vermicompost were Eiseniafotida worms.
Results: According to the results of this study, a significant decrease was observed in the number of fecal coliforms in Sludge- domestic waste; as the number of fecal coliforms reduced from 7500000 (MPN/g), in the raw sample, to 1500 (MPN/g), eight weeks after the outset. Removal efficiency of fecal coliforms was 99.98 percent. Moreover, according to the obtained results, the mixture of Sludge and domestic waste had some parasite eggs (22 number/gr) in the raw samples. This amount was fully removed by the process of vermicomposting, during the forth week.
Conclusion: The results showed that earthworms are a great ability to remove pathogens but to reach the standard set in mixed microbial treatment sludge - waste can not be sure.

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Introduction

The solid wastes are the unwanted organic and non-organic materials generated from various sources such as agriculture, industries, restaurants, and treatment plants. These wastes are usually land filled by incineration, scattering over the earth surface, and burying which can contaminate the air in addition to the water and soil [1]. Fertilizing is the second strategy of the four solid waste management strategies in the modern system of the solid waste management due to its economic and health importance. This is applied for the mixed or separate municipal wastes such as gardening wastes, food wastes, leaves, agricultural wastes, animal wastes, and biosolids. Vermicomposting is a semi-aerobic process (80 % moisture) that is performed by certain species of worms, fungi, bacteria, and actinomycetes. It is also a combination of materials resulted from the worms' growth substrate which remains in the environment after excretion of waste materials from the worm's digestive system. Thus, this material is a collection of the worm's castings as well as decomposed organic materials and worms' bodies which are of great nutritional value for plants [2, 3]. Lack of enough information about the presence or absence of the human pathogens in the wastes' vermicompost is one of the reasons that vermicompost is not widely accepted. Although, vermicomposting of wastes is performed in the thermophilic phase, but it is applied in mesophilic phase, because heat greater than the temperature range of mesophilic phase will lead to the earthworms' death [4]. Various reports showed the changes' trend of the pathogens in vermicomposting, the first of which was presented by USEPA [5]. Additionally, Eastman, reported the decrease of salmonella fecal coliforms in vermicomposting of municipal wastewater biosolids [6]. Xuelian Liu et al, also stated that Escherichia coli O_{157}:H_7 decreased in artificial soil vermicomposting [7]. The results of other studies conducted by Fernando Monroy in 2008 & 2009, presented that densities of the nematodes, protozoa, and fecal coliforms have decreased while wastes were passing through the worm's gut [8, 9]. Moreover, Manuel Aira et al., reported considerable decrease of pathogens in vermicomposting of cow manure [10]. The results of the study conducted by Fernando Monroy on the microbial and chemical properties' changes of the produced fertilizer in the compost and vermicompost processes showed considerable reduction of the microbial agents after 60 days [11]. Yet, in another research, Karimi et al. studied vermicompost process resulted from mixed treatments of the cow manure, sludge of the wastewater treatment plant, and food residues, they entrenched that the fecal coliforms and parasite eggs decreased considerably [12].

Fertilizers produced by vermicomposting are very useful for improving soil structure, but in the case of pathogenic agents' presence, they can cause and transmit diseases. Thus, this study aimed to investigate the efficiency of the vermicompost process in microbiological quality improvement of the produced compost.

Materials and Methods

This experimental study was conducted in the pilot scale in the laboratory of Public Health School, Yazd Shahid Sadooghi University of medical sciences. In order to produce vermicompost, a mixture of the household biodegradable wastes, consisting of the food residues, waste vegetables and fruits, and wastewater treatment plant sludge was used in a reactor with dimensions of 15 × 30 × 50 cm. The pilots' operational requirements included an-eight week period, aerobic condition, and environmental temperature of 30 ± 5 °C. The percent moisture was 70 % and the Eiseniafotida worms were applied for vermicompost production. Two containers were applied for each bed to make air alternation possible and prevent earthworms from escaping. The sampling was carried out during 57 days of the process and repeated twice. A 50 gram sample was taken in each sampling and finally, 68 samples were collected.

The experiments of the microbiological quality determination were performed in the initial stages, during the process, and the final product. These experiments consisted of determining the probable numbers of the fecal coliforms and parasite eggs. Specific culture medium, A_1, was applied for the fecal coliform and Zinc sulfate was used for diagnosing the parasite eggs [13, 14].

After preparing the proposed culture media, 5 g of the sample was inoculated to the enrichment culture medium of peptone water nonselective liquid. Then, after it was placed in the shaker incubator device at 25 °C for 5 min, it was replaced to another incubator at 37 °C for 16-20 hours.

The coliforms were diagnosed by using the 9-tube fermentation and A_1 medium. In the coliform test carried out using multiple tube fermentation, three concentrations of 1, 0.1, and 0.01 were used under serological bain-marie conditions of 41.5 °C for 20-24 hours.

In order to conduct the experiment of parasite eggs' diagnosis, initially the tube was filled with the zinc sulfate solution to the half. Then, the suspension was made by transmition of a 1 g sample. The mentioned suspension was passed through a double-layer cloth filter and the filtered solution was returned to the tube. The zinc sulfate solution was added to the tube until it reached up to 2-3 mm of the tube muzzle. After it was centrifuged at 3000 rpm speed for 1 min, one loop was taken from the upper layer solution of the tube and then it was added to the slide containing the lugols so that the diagnosis process can be performed under the microscope.

In order to study the effect of treatment on the coliforms' number, one-way analysis of variance (ANOVA) were used through the SPSS software, in which treatments were the factors and weeks were set as the blocks. The statistical significance level was set at 0.05. Due to the abnormality caused by the fast growth of data and to perform ANOVA, the mathematical logarithm was applied on the coliforms' numbers.

Ethical issues

This study was conducted with the approval of Shahid Sadoughi University of Medical Sciences and Health Services, Medical Ethics Committee. Code: IR.SSU.SPH.REC.1394.34

Results

On the basis of the results of this study, there was a substantial reduction in the fecal coliforms of the mixed sludge -household wastes, so that the fecal coliform numbers in the raw sample reached from 7500000 (MPN/g) to 1500 (MPN/g) in the eighth week (Figure 1).

Figure 2 shows that the mixture of sludge treatments and household wastes has 22 number/g parasite eggs in raw samples which was completely removed in the fourth week by vermicompost process. The physio-chemical properties of the raw samples after performing the relevant experiments are presented in Tables 1 and 2. These properties consist of moisture, nitrogen, organic carbon, PH, and electrical conductivity (EC). The moisture content of the household wastes is significantly different from those of the cow manure and sewage sludge, but other parameters are slightly different.

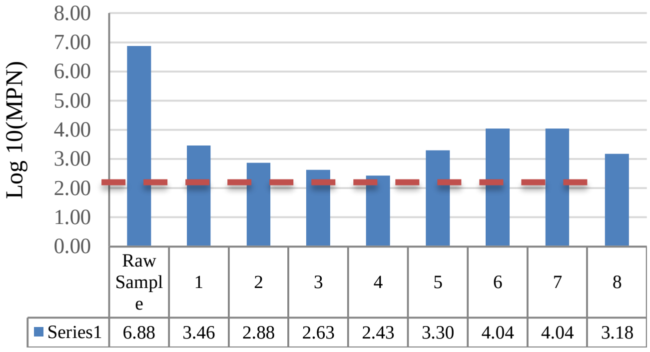

Figure 1: Change trend of the fecal coliforms for sludge-waste treatment (logarithm 3 presented in the diagram is the standard value of fecal coliform in class-A compost of Iran.)

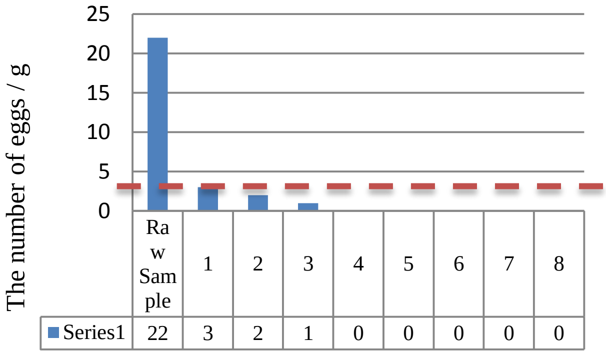

Figure 2: Change trend of the parasite eggs' numbers in vermicompost resulted from the mixed treatments of sludge–wastes during 8 weeks (Number 1 in the diagram is the parasite eggs' standard value in class-A compost of Iran.)

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Table 1: Physio-chemical parameters of the wastewater treatment plant sludge and household wastes in the raw sample.

Parameters	Sewage sludge			Food residuals		
	Mean ± SD	Maximum	Minimum	Mean ± SD	Maximum	Minimum
Moisture (%)	17.25 ± 2.21	20	15	71.85 ± 2.65	73.7	70
EC (mmhose/cm)	2.17 ± 0.69	2.9	1.4	1.459 ± 0.00	1.46	1.46
Organiccarbon (%)	70.26 ± 8.06	79.8	40.24	54.02 ± 0.02	54.16	53.88
Nitrogen (%)	5.41 ± 0.65	7.9	2.1	1.69 ± 0.014	1.7	1.68
pH	7.06 ± 1.09	8.11	5.86	6.3 ± 0.14	6.4	6.2

Table 2: Coliform numbers in the mixed vermicompost resulted from sludge-food residuals on the basis of the different weeks

log_colif

	N	Mean	Std. Deviation	Minimum	Maximum
0.00	2	6.88	0.00	6.88	6.88
1.00	2	3.46	0.00	3.46	3.46
2.00	2	2.86	0.021	2.85	2.88
3.00	2	2.63	0.00	2.63	2.63
4.00	2	2.43	0.00	2.43	2.43
5.00	2	3.31	0.015	3.30	3.32
6.00	2	4.04	0.00	4.04	4.04
7.00	2	4.04	0.00	4.04	4.04
8.00	2	3.18	0.00	3.18	3.18
Total	18	3.65	1.29	2.43	6.88

The one-way analysis of variance showed that the mean of fecal coliform is at least different for one week ($p < 0.001$). The Tukey's test was also applied for further details which is presented as follows: (Table 3)

Table 3: Results of the Tukey's test for diagnosing the differences between the weeks

log_colif
TukeyHSD[a]

Time	N	Subset for alpha = 0.05							
		1	2	3	4	5	6	7	8
4.00	2	2.43							
3.00	2		2.63						
2.00	2			2.86					
8.00	2				3.18				
5.00	2					3.31			
1.00	2						3.46		
6.00	2							4.04	
7.00	2							4.04	
0.00	2								6.88
Sig.		1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 2.00.

Results of the Tukey's test showed that there is a significant difference between the first and the eighth week ($p < 0.001$).

With respect to the descriptive statistics' table on the fecal coliform numbers' logarithm in different weeks, the mean of fecal coliform logarithm was 3.18 in the eighth week placed at the 95% confidence interval. So, at the end of the eighth week, the fecal coliform numbers in the vermicompost of the mixed treatments of sludge-waste did not reach the standard limits of the relevant organizations.

Discussion

Removal of the pathogenic agents from the wastes like the household wastes and the wastewater treatment plant sludge which can be used for the soil amendment, is crucial for preventing transmission of diseases. The vermicompost has the ability to remove the pathogens considerably. Transmission of different diseases through the earthworm's gut affects the existing microbial population of different treatments. Decrease of the pathogens in vermicomposting depends on different factors such as the enzymatic activity of the earthworm gut, secretion of the coelomic fluids with antibacterial properties, and also competition among different groups of microorganisms.

Current study shows that the maximum reduction of the fecal coliform numbers in vermicomposting resulted from sludge-waste, has occurred in the fourth week. Also, results indicated that the parasite eggs in the vermicompost were not diagnosable after four weeks.

The results of study conducted by S.M. Contrerus on “Vermicomposting resulted from biologic wastes with cow manure and oat straw” reflected that the parasite eggs' numbers were not diagnosable after 60 days and the fecal coliforms decreased considerably. The mentioned results are consistent with those of the current research [15]. The results of the study carried out by Monory et al, on the density decrease of nematodes, protozoa, and fecal coliforms caused by passing through the earth worm's gut, showed that the nematodes and fecal coliforms decreased considerably which are in the same line with the results of this study. Decrease in the nematodes' numbers can perhaps be explained in two ways; first, because it is a part of the earthworm's food, second, removal of nematodes and pathogens by proteolyti cenzymatic activity [8]. Further, Monory et al., reported that the coliforms' maximum reduction in the pig slurry was because of the earthworm's transmit in the second week. The mentioned results does not support the results achieved from the vermicomposting of mixed treatments of sludge - household wastes. This can be due to the differences in the treatment types and also the research requirements. Moreover, in this study, the coliforms' numbers have increased after four weeks that is consistent with the results of the present study [9]. LG Rodriguez investigated the reduction of pathogens' numbers in tank's septic sludge in vermicomposting and showed that the pathogens' numbers have decreased considerably which is in the same line with results of the present study [16]. The results of the study carried out by Aira et al, on pathogen decrease in cow manure showed that the fecal coliform reduction has not reached the EPA standards that is consistent with the results of this vermicomposting [10]. Subrataitail reported that in the vermicompost final product resulted from wastewater primary sludge, the pathogens decreased considerably and were less than the standard limits of USEPA. The mentioned results are inconsistent with those of the current study which may be because of the difference in the treatment types used in these two researches [17]. The results of the study carried out by Karimi et al, represented that the fecal coliforms' numbers in the vermicompost resulted from mixed treatments of cow manure-sludge-waste did not reach the set out standards of different organizations. This is in the same line with the results of this study [12].

Conclusion

Results of the current study showed that earthworms have a high ability to remove the pathogens with no need of temperature increase in

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vermicomposting. However, they don't have the necessary ability to reach the determined microbiological standard in the mixed treatments of sludge-waste.

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Conflict of interest

We have no competing interests.

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