ISSN: 1394-7990
Malaysian Journal of Soil Science Vol. 24: 135-146 (2020) Malaysian Society of Soil Science

Study of Microbial Respiration in Different Types of Vermicompost

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ABSTRACT

The respiration of microorganisms in soil is one of the indexes of microbial activity of the soil. However, it is known that soil microbial respiration has decreased due to heavy use of chemical fertilisers and lack of organic matter in the soil. Hence, this study was carried out to measure the effects of different types of vermicompost on microbial respiration. The study was conducted in a completely randomized design with four replications in 2014 in the laboratory of Soil Science, College of Agriculture and Natural Resource, Gonbad Kavous University, Iran. Treatments consisted of different mixtures of vermicompost with soil and without soil that were produced from different manures. The results showed that the type of vermicompost with soil and without soil had a significant effect on microbial respiration. The most and least microbial respiration rates were observed in 50% cow manure + 50% potato and in 25% straw + 75% horse manure, respectively, when tested without mixing with soil. Means comparison of microbial respiration in treatments by 50% composition (vermicompost + soil) showed that the lowest and highest rates of microbial respiration was in 25% straw + 75% horse manure with 735 mg carbon per kg composition and 75% straw + 25% poultry manure at 1934 mg carbon per kg composition respectively. Our study found that vermicompost increased the organic matter of soil and created a good environment for microorganisms to live in the soil.

Key words: Microbial respiration, cow manure, sheep manure, palm, alfalfa.

INTRODUCTION

The sustainability of agricultural systems is considered an important issue around the world. Disturbance of biological activity, reduction of biological nitrogen fixation and other nutritive elements have increased by frequent application of chemical fertilisers for agriculture. It is necessary to gradually reduce the use of chemical fertilisers and substitute it with organic fertilisers especially vermicompost (Palm *et al.* 2001). Vermicompost benefits the environment by reducing the need for chemical fertilisers and decreasing the amount of waste going to landfills. It also improves soil aeration, microbial activity, water holding capacity and nutrient recycling.

Soil respiration is one of the oldest parameters to determinate microbial activity in the soil (Kieft *et al*. 1987). It is one of the most important soil fertility

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indicators, which activity requires energy (Nanipieri *et al.* 1990). Therefore, respiration is an overall process that is not confined to microorganisms but is performed by all creatures living in soil. The humidity, temperature, ability to access nutrients (Ramnarain and Ansari 2019), carbon storage (Fang and Moncrieff 2005), vegetation type, quality and quantity of substrate, biomass and microbial activity, management and land use especially fertilizer consumption are among the factors that affect respiration (Ding *et al*. 2007; Biabani *et al*. 2018). Animal manure and vermicompost have been used as fertilisers since centuries ago.

Poultry manure which has a high amount of nitrogen has now emerged as one of the best organic fertilisers (Sloan *et al.* 2003). Fereidooni Naghani (2009) found that using two types of fertilisers (urea and poultry fertilizer) increased soil CO_2 during its use, with poultry manure increasing CO_2 much more than urea due to its ability to access carbon and essential nutrient quantities. Min *et al*. (2003) also reported the increase in basal respiration in treatments under cow manure compared with chemical fertilisers in the alfalfa fields. Various factors, such as amount of carbon, access to nitrogen, moisture content, temperature, ventilation and worm population, emissions of CO_2, CH_4 and N_2O, influence vermicompost production process (Hobson *et al*. 2005).

Many researchers have shown that soil organic matter is a source of energy and food for microorganisms. The quantity and quality of organic matter are among the factors that have an influence on exacerbating soil biological activities (Palm *et al.* 2001). Thus, for greater productivity of plant residue in agricultural systems, understanding the process of analysing plant residue, factors affecting analysis of materials and speed of releasing nutrients is necessary and important (Palm *et al*. 2001). Several studies have shown that the speed of decomposition and mineralization of plant materials depend on environmental conditions and chemical composition of crop residue (Kieft *et al*. 1987; Martens2000; Bossuyt *et al*. 2001; Palm *et al*. 2001). In terms of differences in the compounds such as protein, cellulose, pectin, hemicellulose and polyphenols, a compound of plant species in an ecosystem has a significant effect on organic matter decomposition rate, the cycle of nutrients and finally soil fertility status (Martens 2000; Bossuyt *et al*. 2001; Palm *et al*. 2001; Hadas *et al.* 2004). During the process of producing vermicompost, earthworms break down organic matter. Moreover, the added mucus and enzymes increase microbial activity of earthworms and during this process microorganisms also degrade organic carbon through release of CO_2. Sothar (2006) reported that earthworms cause loss of organic carbon in the process of vermicomposting due to microbial respiration. Garg *et al*. (2005) reported a 58.4% reduction in organic carbon in cow manure and 55.4% in equine manure for 90 days during the vermicomposting process.

Sinha and Heart (2012) argue that organic material convert into vermicompost. This, in turn, can cause the entry of a great deal of atmospheric carbon which is absorbed by photosynthesis. Furthermore, it can improve soil fertility and prevent greenhouse gas emissions into the atmosphere that occurs through burning or abandoning organic matter in nature.

Borken *et al*. (2002) conducted a study on changes in microbial and soil properties following compost treatment of degraded temperate forest soils. They state that any increase in temperature and use of compost can give rise to an increase in soil microbial biomass carbon and respiration. Tejada *et al*. (2006) state that an increase in microbial biomass carbon is generally related to positive effects of organic fertilisers in the soil. In addition, the simple materials which provide organic fertilisers stimulate microbial and enzyme activities in the soil.

Since soil microbes are responsible for decomposition, the type of plant residue in terms of differences in the amount and type of organic compounds (quality of substrate) have a great impact on different soil biochemical processes (Pascual *et al.* 1999). A study on atmospheric residue (biomass burning) which has been utilized, show that the residue can be a substrate containing considerable amounts of carbon and can be used as a source of energy (Bautista *et al.* 2011). On the other hand, since the ratio of carbon to residual atmospheric nitrogen is greater than 30, the mix of these materials with soil makes nitrogen organic. Thus, microbial respiration in soil treated with plant residues shows a significant increase compared to pure soil (Nourbakhsh 2004). Other studies have shown that when plant residue is added to the soil, the emission rate of carbon dioxide expresses the respiration activity of soil microorganisms because of a significant increase in the decomposition of organic matter that is added to the soil (Martens 2000; Hadas *et al.* 2004). The application of plant residue to the soil not only affects soil chemical and biochemical properties, but also affects the rate of conversion of nutrients into usable forms for plants.

C/N is among the biochemical characteristics of an organic material that has a great influence on its decomposition rate. A material with lower C/N ratio, like soybean residue, compared to a corn residue which has a higher C/N, for example, would be decomposed more easily by microorganisms (Manzoni *et al.* 2008.)

This study was conducted with the aim of investigating the effect of different types of vermicompost on soil microbial biomass respiration, in relation to the importance of organic compounds in the health of agricultural and non-agricultural ecosystems.

MATERIALS AND METHODS

The study was carried out in Gonbad Kavous University (Iran) in 2014 with four replications under laboratory conditions in two completely randomized designs. The treatments were normal soil (control) and vermicompost made from 100% cow manure, 75% cow manure + 25% potato, 50% cow manure + 50% potato, 75% cow manure + 25% cabbage, 50% cow manure + 50% cabbage, 75% cow manure + 25% palm leaf, 50% cow manure + 50% palm leaf, 25% cow manure + 75% palm leaf, , pure straw, 50% straw + 50% sheep manure, 25% straw + 75% sheep manure, 75% straw + 25% poultry manure, sheep manure, 25% alfalfa +

TABLE 1
Amount of C/N in vermicompost

Treatment	C:N
Oil	9.71
100% cow manure	11.42
75% cow manure + 25% palm leaves	11.01
50% cow manure + 50% palm leaves	10.69
25% cow manure + 75% palm leaves	11.03
75% cow manure + 25% potato	10.33
50% cow manure + 50% potato	12.88
25% cabbage +75% cow manure	14.94
%50 cabbage+ 50% cow manure	10.69
Net straw	16.82
Horse manure	13.84
Sheep manure	11.89
50% horse manure + 50% straw	14.26
25% straw + 75% horse manure	13.98
50% straw + 50% sheep manure	12.77
25% straw + 75% sheep manure	12.42
25% alfalfa + 75% sheep manure	11.47
50% alfalfa + 50% sheep manure	11.76
75% straw + 25% poultry manure	11.84

75% sheep manure, 50% alfalfa + 50% sheep manure, horse manure, 50% straw + 50% horse manure, 25% straw + 75% horse manure (Table 1).

In addition to the above mentioned treatments, another set of compound vermicompost mixed with soil in the ratio of 50:50 was prepared. In order to measure microbial respiration, 50g of different types of vermicompost and 100g mixture of vermicompost with soil from each treatment were placed in one liter plastic containers. In order to retain soil moisture content, distilled water equal to 65% of field capacity was added. Dishes were placed in an incubator at a temperature of about 25-30°C (lids of plastic containers were closed in order to prevent entry and exit of gases). The carbon dioxide (CO_2) resulting from microbial respiration was collected in 0.5 N NaOH. To do this, 10 mL of 0.5 N NaOH was poured into a small can (film canister) and was placed on the surface of the vermicompost. The microbial respiration of vermicompost was titrated with 0.5 N HCl for a duration of 33 days as shown in Table 2. Before titration, 5 mL, 10%

TABLE 2
Titration time

Titration row	Week days	Duration (days)
1	First week	3
2	First week	3
3	Second week	4
4	Third week	5
5	Fourth week	6
6	Fifth week	6
7	Sixth week	6

barium chloride ($BaCl_2$) was added so that carbonate ion (CO_3^{-2}) was precipitated in the form of barium carbonate ($BaCO_3$) and the remaining NaOH was titrated with 0.5 N HCl. The container without vermicompost (pure soil) was considered as a control by adding NaOH. The C/N of treatments was also measured (Table 1). The results of this study were analysed using SAS statistical software and the difference between means was calculated using LSD.

The total reactions that occurred in this experiment were as follows:

$$CO_2 + 2NaOH \longrightarrow Na_2CO_3 + 2H_2O$$
$$Na_2CO_3 + BaCl_2 \longrightarrow BaCO_3 + 2NaCl$$

The remaining amount of NaOH was determined with titration by HCl. Finally, the amount of CO_2 caused by microbial respiration was calculated as follows:

$$CO_2 \text{ or C (mg/kg)} = (B-S) \times N \times E$$

Where

B= mL of acid used for control
S= mL of acid used for sample
N= normality acid
E= the highest equivalent weight (22 for CO_2 and 6 for C)

Since the amount of used vermicompost was 50g and 100g, to convert it into one kg, the calculated value was multiplied by 20 and 10, respectively so that the amount of respiration was calculated and reported based on mg carbon in kg soil.

RESULTS

As shown in Table 3, the use of different types of vermicompost of animal manure and plant residue in composition with soil and non-blending with soil had a significant effect on microbial respiration at 1% probability level.

TABLE 3
Analysis of variance for microbial respiration

Source of changes	Degree of freedom	Mean squares	
		Microbial respiration without soil	Microbial respiration with soil
Vermicompost	18	3275191.26**	519308.89**
Error	57	12427.74	699.03
CV (%)	-	4.75	2.04

** Significant at 1% probability level

Organic matter is not just one compound. The C compound present in soil and vermicompost is basically different in its chemical nature and structure. Notably, in order to arrive at a conclusion on the chemistry of the compound, just imagine the case where the lignin or chitin carbon is similarly treated with the sugar or cellulose or starch carbon. Decomposition is a process that greatly depends on the activity of microorganisms and the nature of organic C and its vulnerability towards microbial activity. Hence, the structure and nature of carbon compounds in soil and vermicompost need to be ascertained first, before a real picture on decomposition can be assessed (Chudek 2008).

The mean comparison of microbial respiration in treatments without soil and microbial respiration with soil showed a significant difference in microbial respiration between treatments without soil and treatments with soil (Table 4). Microbial respiration in treatments without soil (2349 mg carbon in kg soil) was more than in treatments mixed with soil (1292 mg carbon in kg soil), almost two times greater (Table 4).

Table 5 shows the mean of microbial respiration in different types of vermicompost in comparison with control, mixed with soil and without. In the treatment where it was mixed with soil, microbial respiration rate was variable, from 336 mg carbon per kg of soil (normal soil) up to 3829 mg carbon per kg of composition. The maximum rate of microbial respiration was obtained from 50% cow manure + 50% potato treatment, while 25% straw + 75% horse manure treatment revealed minimum rate (1404 mg C/kg of microbial respiration).

TABLE 4
Comparison of microbial respiration with soil and microbial respiration without soil in treatments under study

Treatment	Mean	SEM	t	Pr> t
Microbial respiration without soil	2349.24	40.58	31.58	0.0001
Microbial respiration with soil	1292.41	102.30	22.96	0.0001

TABLE 5
Comparison of mean of microbial respiration (mg carbon per kg of soil)

Treatment	Treatment levels	Microbial respiration without soil	Microbial respiration with soil
Vermicompost	Normal soil (control)	336.00k	336m
	100% cow manure	3208.50c	1319.25f
	75% cow manure + 25% palm leaves	2595.00e	1389.75e
	50% cow manure + 50% palm leaves	2970.00d	1522.50c
	25% cow manure + 75% palm leaves	2697.00e	1481.25d
	75% cow manure + 25% potato	3250.50c	1545.74c
	50% cow manure + 50% potato	3829.50a	1662b
	25% cabbage +75% cow manure	3334.50c	1520.25c
	%50 cabbage+ 50% cow manure	2416.50f	1375.50e
	Net straw	2901.00d	1482d
	Horse manure	1567.50i	1249.50gh
	Sheep manure	1813.50gh	1473.75d
	50% horse manure + 50% straw	1693.50hi	1282.50fg
	25% straw + 75% horse manure	1404.00j	735 l
	50% straw + 50% sheep manure	1831.50gh	1123.50i
	25% straw + 75% sheep manure	1860.00g	901.50k
	25% alfalfa + 75% sheep manure	1777.50gh	1003.50j
	50% Alfalfa + 50% sheep manure	1590.00i	1218h
	75% straw + 25% poultry manure	3559.50b	1934.25a
	LSD (0.05)	157.85	37.43

Means within each column with at least a letter in common are not significantly different at α= 0.05

Comparing the average of microbial respiration in the treatments in combination with soil, a significant difference was found between treatments. Treatments of 25% straw + 75% horse manure treatment with 735 mg carbon per kg of composition and 75% straw + 25% poultry manure treatment with 1934 mg carbon per kg of composition had the lowest and highest rates of microbial respiration, respectively(Table 5).

Comparing the microbial respiration activity of vermicompost of different types of animal manure and plant residue mixed with soil and without soil (Figure1), compost treatments in combination with soil had less microbial respiration than treatments without soil. Microbial respiration changes were uneven in the treatments, but as shown in Figure 1, in every treatment, changes in microbial respiration without mixing in soil showed a relatively constant increase compared to the same treatment in combination with soil.

DISCUSSION

The application of plant residue and different types of vermicompost in the soil not only affects soil chemical and biochemical properties, but also the decomposition rate of conversion of nutrients into usable forms for plants. Therefore, any process that influences soil biological populations will lead to some changes in soil enzyme systems. We can say that the soil-enzyme systems are interconnected with the management of organic waste materials (Michael and Joshua 2003). It appears that treatments without mixing with soil provided a more appropriate environment and sources of energy for microorganism activity. One of the reasons for the decomposition of organic matter is the presence of a lot of available nitrogen as well as some toxins that exacerbate microbial activity (USDA 2003). One of the reasons for low microbial respiration in 25% straw treatment + 75% of horse manure was its low nitrogen content (C/N =13.98).

There is a considerable increase in carbon mineralization rate, respiration and microbial biomass of the compost because of the composting process itself. It is possible that the high rate of inorganic carbon in treatments containing organic matter (compost) was due to moisture content and an increase in microbial activity which resulted in increased readily available and easily analysable energy source for microbes.

Differences in the impact of various organic substances on the amount of microorganisms in the whole soil are likely to be related to the complexity of their chemical structure and their ability to supply carbon and other nutrients required for soil microorganisms, as well as the C/N ratio. The chemical composition or quality of organic matter or the amount of organic matter lignin, microbial population structure, soil moisture, availability of nutrients, salinity and soil structure are important factors that have a significant effect on the degradation of plant remains or microbial respiration. There could be several reasons for the difference in microbial respiration in the different treatments. The amount of lignin present in the naturals used for treatments is likely to vary. Speratti *et al*. (2007) reported that earthworms influence the emission of CO_2 from the soil ecosystem. The measurement of respiratory carbon dioxide caused by metabolic activity and enzymes is a common indicator of compost maturity.

Fereidooni Naghani (2009) observed that within a few weeks of animal manure incubation consumed by earthworms, respiration and enzyme activity during this period was greatly reduced. It was also observed that as time progressed, microbial activity increased due to addition of urea and poultry manure. The researchers found that the lowest rate of decomposition among organic fertilisers was related to materials that had undergone composting (Morank *et al*. 2005). Perhaps some treatments, such as cow + potato fertiliser, have a lesser composting duration but have an improved composting process, in contrast to manure + straw. Speratti *et al.* (2007) believe that earthworms through feeding and replacement of materials can increase respiration in aerobic microsites. In a laboratory scale study, Atiyeh et al. (2000) found that earthworms cause biochemical changes in cow manure leading to accelerated maturity; this makes the organic waste material stable with an increase in total nitrogen without large changes in acidity.

When used the amount of straw for making vermicompost increases respiration. This increase is due to having large amounts of carbon and its easily decomposable analyzable compounds and supply of energy for microorganisms in the soil, Microorganism activity increases and CO_2 sublimation rate increases during respiration. Chicken manure with higher levels of nutrients, in comparison with other treatments, increased the microbial population. Therefore the higher amount of organic matter dispersed in the soil, resulting in higher available substrate for use of soil micro-organisms, increased the CO_2 outflow rate of soil. This will result in an increase in microbial activity and living organisms present in the soil which will lead to an improvement in the chemical and physical property complex of the soil. It is recommended that further studies be carried out on the effects of different ratios of different vermicompost on soil microorganism life.

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