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Different Substrates on the Reproduction Rate of Earthworm (*Eudrilus eugeniae*) and NPK Content of Its Castings

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ABSTRACT

In response to the posing threat of chemically produced crops using synthetic fertilizers for human consumption, organic farming using earthworms in composting has resorted. This study is carried out to determine the effects of locally available substrates on the reproduction rate of earthworms and the NPK content of its castings. It was laid out with seven treatments replicated three times in Completely Randomized Design. Treatments used were 75% goat manure plus 25% of various substrates such as banana leaves, grass clippings, rice straw, legume leaves, cassava leaves, banana bracts, and sawdust. They were shredded, partially decomposed, transferred to the beds for earthworm feeding, and harvested after 45 days. The castings were collected, separated from the biomass, weighed, and were sent for laboratory analysis. The highest reproduction rate of earthworm biomass was in Treatments with sawdust and legume leaves. Treatments with banana bracts and legume leaves had significantly higher Nitrogen and Phosphorous. Higher Potassium content was noted in treatments with sawdust and legume leaves. It is recommended to use 75% goat manure and a combination of legumes leaves and sawdust for a higher rate of reproduction of vermi biomass and higher contents of Nitrogen, Phosphorous, and Potassium (NPK) in the castings.

Keywords **—** Agriculture, Substrates, biomass, reproduction rate, C: N ratio, vermicast, Philippines

INTRODUCTION

The idea of using worms to break down the organic waste is not new. In recent years, there is an awakening about the importance of earthworms in degrading bioorganic wastes. They are significant in waste recycling, especially urban and industrial wastes. Hence, earthworms are known to be multipurpose organisms. The breakdown of organic waste by the ingestion of earthworms is vermicomposting, which transforms the material into worm castings and is considered an excellent form of organic fertilizer (Domínguez, Edwards, & Webster, 2000). Researchers contend that the microorganisms present in the gut of worms aid in decomposition and provide nutrients for plant growth and development. Instead of burning wastes, which can eventually contribute to ozone depletion and global warming, organic wastes could be transformed into viable, environmentally sound, and healthy farm inputs in the form of organic fertilizers, the vermicast and vermicompost.

There are two major practices involved in this process, vermiculture and vermicomposting. In vermiculture, the aim is to continuously increase the number of worms to achieve a sustainable harvest, while in vermicomposting, one would want to keep the population density low so that reproductive rates are optimized to grow worms. The more worm produced, the more castings and compost generated.

Vermicomposting can help protect the environment, enhance and maintain soil fertility, and create livelihood opportunities for rural families through the ample supply of material in farms. The worm *per se* called the Biomass commands higher prices per kilo. It is also used as a substitute for protein sources in feeds for animals as in livestock and fishes. It has been proven to have higher protein content compared to fishmeal. The castings and compost as residuals in vermiculture are other sources of supplemental income for farmers.

Vermimeal production, on the other hand, can be considered the most economically feasible application of vermiculture, considering the growing demand for animal feeds aggravated by the continued growth of the human population. Vermiculture has bright opportunities in the livestock feed industry, according to Kale, as cited by Adhikary (2012). A feed preparation that consists of processed earthworm biomass is vermimeal or earthworm meal. For animals, birds, and fishes, it is a rich source of protein, amino acids, fats, vitamins, and minerals. To produce a kilogram of vermimeal, about 5.5 kg of fresh African Night Crawlers (ANC) biomass (18 percent dry matter) is required. It can be wrapped in plastic bags and stored for up to 3 months in a cold, dry place. The following composition was discovered in an estimated study of an ANC vermimeal in dried and pulverized form; 68 percent crude protein, 9.57 percent fat, 11.05 percent ex-tract nitrogen-free, and 9.07 percent ash. Numerous researches on numerous species of cattle, birds, and fish traced its foundation because these earthworm species are truly the food for wild animals in the forest.

In a study conducted by the Agricultural Training Institute (ATI) Philippines, they found out that vermicomposting enhances the texture of the soil and increases the soil's water-holding ability. It can be low in NPK, but it provides important nutrients not present in inorganic fertilizers such as calcium, magnesium, manganese, copper, iron, and zinc. Also, it has microbial activities that foster plant health and resistance to pests/diseases.

Among the beneficial uses of vermicompost includes: Red worm castings have a higher concentration of humic substances (Adhikary, 2012). Humus allows the aggregation of soil particles into clusters, establishing air passage channels and increasing its water-retaining ability. The presence of worms regenerates compacted soils and increases over 50 percent water penetration in those soils. Data revealed that 10,000 worms in a field plot have the same value as three farmers who work 8 hours of shift during the year with 10 tons of manure throughout the plot. For the plant nutrients, humic acid found in humus supplies binding sites such as calcium, iron, potassium, sulfur, and phosphorus. These nutrients are retained in a form readily available to plants in humic acid and released when needed by plants. Humic acid in vermicompost promotes the development of plants even in small quantities. For plants, humic acid in the humus is important in four fundamental ways: (a) Allows plants to absorb nutrients from the soil; (b) Helps to remove unresolved minerals in the soil; (c) Stimulates the growth of roots, and (d) Helps to overcome stress in plants.

Humus found in vermicast and composts helps prevent dangerous plant parasites, fungi, nematodes, and bacteria (Adhikary, 2012). Vermicompost can combat diseases of soil-borne plants such as root rot. Humus also enhances water permeability and water-retaining capacity, leading to increased plant health and soil moisture usage more effectively. The abundance of nitrogen in vermicompost is observed to be greater than in aerobic compost piles. Other agronomic advantages of the application of composts, such as an elevated rate of suppression of soil-borne diseases and soil salinity reduction, exist. One research recorded a reduction in mean root disease in tomatoes from 82 percent to 18 percent and capsicum in soils adjusted with compost from 98 percent to 26 percent. It was also found out that plants fertilized with vermicompost can establish biological resistance in plants, the ability to minimize pests attack, and the ability to suppress plant disease. Vermicompost contains some antibiotics and actinomycetes that increase the power of biological resistance against pests and diseases among crops. When earthworms and vermicompost were used in agriculture, spraying of chemical pesticides was dramatically decreased by over 75 percent.

A worm cast (also referred to as vermicast) is a biologically active mound containing thousands of plant substance bacteria, enzymes, and remains that have not been digested by worms (Adhikary, 2012). Microbial production in worm castings of beneficial microorganisms is ten to twenty times greater than in soil and other organic matter. Nitrogen-fixing & phosphate solubilizing bacteria, actinomycetes & mycorrhiza fungi are among the beneficial soil microbes activated by earthworms. Studies showed that more than 10/gm of bacterial count in sample vermicompost. Actinomycetes, Azotobacter, Rhizobium, Nitrobacter & Phosphate solubilizing bacteria were found in 102 to 106 per gm of vermicompost.

Castings contain nutrients that are slowly released and readily accessible to plants (Adhikary, 2012). The plant nutrients that are encased in mucus membranes secreted by the earthworms are found in castings. Rather than permitting rapid nutrient leaching, they melt steadily. The substance has outstanding soil composition, porosity, and capabilities for aeration and water preservation. Castings are capable of carrying 2 to 3 times more water than their soil weight. Root mechanisms are not burned from worm castings. The substance is capable of insulating plant roots from high temperatures, minimizing drought, and weed suppression. It is odorless and consists of products that are 100% recycled. Because of humus material, Vermicompost also has very high porosity, aeration, drainage, and water holding capacity relative to traditional compost.

Worm intestinal behavior is analogous to a miniature composting tube that combines the conditions and inoculates the residues (Adhikary, 2012). For a synergistic interaction and then a terrific byproduct, moisture, pH, and microbial fauna in the gut are favorably preserved. Every day, they swallow vast soil quantities with organic materials (microbes, plant & animal debris), ground them in their gizzards, and eat them with enzymes in their intestines. In the shape of fine mucus-coated granular aggregates called vermicastings rich in NKP (nitrates, phosphates, and potash), micronutrients, and beneficial soil bacteria, only 5 - 10 percent of the chemically digested and consumed material is absorbed into the body, and the remainder is excreted.

For greenhouses or houseplants, as well as planting and farming, worm castings are the finest imaginable potting soil (Adhikary, 2012). Even the most fragile plants will not be burnt, and all nutrients are water-soluble, rendering it an instant-eaten plant meal. In addition to their use as a potting ground, earthworm castings may be used as a planting ground for trees, vegetables, shrubs, and flowers. They should be used as mulch, meaning that the products leach directly onto the earth when watered.

Research indicates that even though plants are already providing “optimal nutrition,” vermicompost usage further enhances plant growth (Adhikary, 2012). It has consistently enhanced the germination of seeds, increased the growth and production of seedlings, and greatly increased plants’ productivity even more than would be possible by the simple conversion of mineral nutrients into usable types of plants. Some studies have also documented that vermicompost produces growth that encourages “auxins,” “cytokinins,” and “gibberellins,” a flowering hormone secreted by earthworms.

A plant bioassay system was used to assess the growth promoter of vermicompost activity. The length of the plumule of seedling maize (Zea mays) was measured 48 hours after soaking in vermicompost water and natural water. The marked difference in plumule length of maize seedlings suggested that hormones that stimulate plant growth are present in vermicompost.

(CSIRO, June 2016) Australia’s Glasshouse studies showed that earthworms composts increased wheat crop growth (Triticum aestivum) by 39 percent, grain yield by 35 percent, grain protein value increased by 12 percent, and crop disease tolerance compared to control.

The results of studies on the development of important vegetable crops such as tomatoes (Lycopersicum esculentus), eggplants (Solanum melangona), and okra (Abelmoschus esculentus) were very strong (Najar, I. A., Khan, A. B., & Hai, A., 2015).

Another research was performed on earthworms' growth effect on okra (Abelmoschus esculentus) as compared to cow manure and chemical fertilizers. Vermicompost has encouraged excellent growth with more flowers and fruits in the vegetable crop. But a significantly lower occurrence of "Yellow Vein Mosaic," "Color Rot," and "Powdery Mildew" diseases was noted in plants applied with vermicompost. It was even considered as one of the most critical discoveries (Adhikary, 2012).

There are an estimated 4,000 earthworm species worldwide, and some 29 species have been identified and found in the Philippines. Earthworms are harmless segmented invertebrates, soft-bodied with no special coverings on their skin wherein their breathing organs are situated. They fed on moist organic materials rich with fungi and bacteria. They are hermaphroditic and parthenogenetic.

Earthworms are classified into three based on their behavior and habitat (Sherman, 2018). The first classifications are the Anecic (Greek for "out of the earth"). These are burrowing worms that come to the surface at night to pull food back into their permanent burrows deep within the soil's mineral deposits. An example is the African Night Crawlers. The second is the Endogeic (Greek for "under the earth"); they are often burrowing worms, but their burrows are usually shallower. They feed on the organic matter situated already in the soil, so they only occasionally come to the surface. Epigeic (Greek for "upon the earth"), these worms live and feed on rotting organic matter in the surface litter. They may not have permanent burrows. The type of worm used in vermicomposting is these "decomposers."

The African Night Crawler (Eudrilus eugeniae) is the most popular earthworm species used in vermicomposting (Ansari & Ismail, 2012). Biodegradable materials like leaves, grass cuttings, manure, and vegetable trimmings are nutritious and suitable food for earthworms. These organic wastes are called substrates or food for worms. The earthworms feed on the substrate and, with the aid of microorganisms in their gut, produce casting deposited on the beds' surface. Their castings contain nutrients that are beneficial to the soil.

A proper combination of carbonaceous and nitrogenous (C: N) organic materials makes an ideal compost or an effective and complete fertilizer. The C:

N ratio relates to the sum of carbon (carbohydrate) and nitrogenous materials needed for energy and development by microorganisms to be composted. The ideal ratio is 25:1 (Guerrero, 2010).

Several studies have been conducted to test the efficiency and effectiveness of earthworms in plant growth and development. The good news is that it eventually shows itself in an increase in yield.

The conversion of organic waste into useful products is essential in recycling organic matter to sustain soil fertility and avoid environmental pollution. Sustainable crop production demands the use of fertilizers because of the loss of soil fertility.

In response to the widely alarming issue of natural resource depletion, pollution, environmental degradation, climate change, and the posing threat of chemically produced crops and animals for human consumption, the government designed a Strategic framework to solve these problems; the Republic Act 10068 or known as "The Philippine Organic Agriculture Act of 2010". It is an act promoting the tradition of organic agriculture in the Philippines that will cumulatively condition and enrich the fertility of the land, increase farm productivity, mitigate emissions and degradation of the environment, avoid the loss of natural resources, further protect the health of farmers, customers, and the general public, and save on imported farm inputs.

One of the pillars of the organic agriculture act is sustainable agriculture. An identified entry point along this field is vermicomposting technology. Guerrero (2010) introduced the importance and viability of earthworm (Eudrilus eugeniae) in the country. Research trials and findings have emerged. Studies on vermicompost production using pig manure were established in the early '80s by Guerrero et.al.

In a study conducted by Cruz in 1996 using Grasses, Pig Manure, Grass + Pig Manure and Grass + Kakawate leaves, it was found out that Grass(75%) + Kakawate (24%) showed significantly higher nitrogen content in the vermicasts compared to the rest of the treatments.

Guerrero (2010) found out that a proper combination of carbonaceous and nitrogenous organic materials makes ideal compost or an effective and complete fertilizer. Furthermore, because some organic materials contain harmful mycelia, antibiotics, plant pests, and excessive heavy metals, proper mixing and composting, leading to dilution and sterilization, may greatly promote the quality of organic materials. Vermi cast was also noted to have stabilized pH regardless of the pH of the substrate.

It was believed that the nutrient composition of the casting has a positive relationship with the substrates that the worm fed in (Edwards, Dominguez, & Neuhauser, 1998). Several studies have revealed to support the positive results of vermicast and composts used for plant growth and development but studies on which substrates fed to worm could give the highest nutrient content in terms of N, P & K is limited. Furthermore, the aim of exponentially increasing the vermi biomass could have been hypothetically affected by the type of substrates fed to the worms, hence this study. Moreover, investigation on the role of substrate fed to earthworms in the nutrient composition of vermicastings and the reproduction rate of earthworm in the local condition is important for a wise decision on the farmers' part to reduce labor cost while increasing productivity in the farm.

OBJECTIVES OF THE STUDY

This study looked into the effects of different substrates on the rate of reproduction of vermi biomass and the NPK content of its casting. Specifically, it aims to (1) Determine which substrate fed on earthworms gives the highest yield of earthworm biomass and reproduction rate, and (2) Evaluate which vermicast has the highest NPK content.

MATERIALS AND METHODS

Research Design

The Completely Randomized Design was used in the study. There were seven (7) treatments that were replicated three (3) times, making a total of 21 experimental plots or vermi beds. Plots were randomized using the randomization procedure of CRD. The farm wastes available in the locality were used as a substrate for the production of vermi added with goat manure in the following specific combinations;

T1 - 75% goat manure + 25% banana leaves;
T2 - 75% goat manure + 25% grass clippings;
T3- 75% goat manure + 25% rice straw;
T4- 75% goat manure + 25% legume leaves;
T5- 75% goat manure + 25% cassava leaves;
T6- 75% goat manure + 25% banana bracts;
T7 - 75% goat manure + 25% saw dust.

Research Site

This trial was conducted in the Experimental Sites of the College of Agriculture and Forestry of the Jose Rizal Memorial State University, Tampilisan, Zamboanga del Norte.

Sourcing out of Vermi Biomass

The earthworms used in the study were sourced out from the Municipal Agriculture Office. They were identified and classified to belong to the same species, the African Night Crawlers (ANC) *Eudrilus Eugeniae,* regardless of age. Biomass was weighed prior to transferring to the beds.

Preparation of Substrates and Beds

Organic materials were gathered and shredded manually and transferred to the suitable beds to allow them to undergo anaerobic decomposition for at least 15 days. The shredded organic materials were covered with a plastic sheet to minimize aeration. As soon as the temperature of the substrate is lowered, the partially decomposed organic materials were transferred to the vermicomposting bed following the ratio of the treatments stated. The beds were measured 1 x 1 m with the substrate at 100kg/bed. Stocking of earthworm biomass was done at 1 kg/bed.

Harvesting and Data Gathering Procedure

After 45 days from the stocking of ANC, harvesting was done manually, segregating the casts and earthworms from vermicompost. Vermicasts settling at the upper portion of the bed were collected for laboratory analysis. The earthworms were weighed and transferred into a suitable medium for future use.

The rate of reproduction was presumed to be the increase in the weight of the biomass before and after stocking. The initial weight of 1 kilogram of vermi biomass was stocked in each bed. The reproduction rate of earthworms was obtained by deducting the initial weight from the final weight of worms divided by the final weight multiplied by 100.

The newly harvested vermicompost was air-dried for 3-4 days, and this was sieved and passed through a 5-mm mesh. Materials that were not passed through were returned to vermicompost bins for the next production cycle.

Chemical Analysis of Vermi casts

The vermicast samples were sent to the Soil Testing Laboratory, Cagayan de Oro City, for chemical analysis to determine the NPK contents.

Statistical Analysis

The data on the weight of biomass or rate of reproduction and NPK content were analyzed based on the Analysis of Variance of the Completely Randomized Design (CRD) procedures. To determine the significance of the different treatments, Duncan's Multiple Range Test was used to determine significant differences between treatment means.

RESULTS AND DISCUSSION

Weight of Earthworm Biomass

Table 1 shows the average weight of biomass after 45 days of the stocking. Analysis of Variance revealed significant differences among treatments. Treatment 7 (75% goat manure + 25% sawdust) and Treatment 5 (75% goat manure + 25% cassava leaves) had significantly higher weight of biomass than Treatments 4 (75% goat manure + 25% legume leaves), Treatment 6 (75% goat manure + 25% banana bracts), and Treatment3 (75% goat manure + 25% rice straw). No significant differences were observed between Treatments 7, 5, 1, and 2 and between Treatments 1, 2, 4, and 6.

The significantly lower weight of earthworm biomass of Treatment 3 (75% goat manure + 25% Napier grass) supports Guerrero's (2010) finding, which found out that vermi beds with graminaceae substrates and rice hulls dramatically lowered down worm population in Nueva Ecija. It could then be concluded that the texture and fibrous composition of these plant species decreases the delight of the earthworms to feed on.

It was reported that vermis was found to exponentially increase in substrates with goat manure. This conforms to the report of Domínguez, Edwards, and Webster (2000) that goat manure resulted in a higher rate of reproduction of biomass due to a lesser amount of metallic elements and stable pH value, which were found to be preferable by vermis.

Reproduction Rate of Earthworm

Table 1 shows the reproductive rate of earthworm biomass. Analysis of Variance revealed significant differences among Treatments. Treatment 7 (75% goat manure + 25% sawdust) and 5 (75% goat manure + 25% cassava leaves) had significantly higher rate of producing biomass than Treatments 4 (75% goat manure + 25% legume leaves), 6 (75% goat manure + 25% banana bracts), and 3 (75% goat manure + 25% rice straw). No significant differences were observed between Treatments 7, 5, 1, and 2 and between Treatments 1, 2, 4, and 6.

Table 1. Rate of Increase in the final weight of earthworm biomass after 45 days of stocking

TREATMENTS	Initial weight of Biomass (g)	Final Weight of Biomass (g)	Rate of Increase in the weight of Biomass (%)
T1	1000	1771.00[ab]	77.10[ab]
T2	1000	1612.75[ab]	61.27[ab]
T3	1000	1240.25[c]	24.00[c]
T4	1000	1400.50[b]	40.00[b]
T5	1000	2143.60[a]	114.36[a]
T6	1000	1352.83[b]	35.28[b]
T7	1000	2149.72[a]	114.97[a]
F-test		**	**
CV (%)		19.16	15.60

Note: Means of the same superscript are not significant at 0.05 level of significance.

NPK Contents of the Castings

Nitrogen (N) Content

The Nitrogen Content of Vermi Cast was found to be significant (Table 2). Treatments 1, 2, and 6 had significantly higher nitrogen content than Treatment 7. However, no significant differences were observed between Treatments 1, 2, 3, 4, 5, and 6 and between Treatments 3, 4, 5, and 7.

This implies that Treatments with 75% goat manure + 25% banana bracts, 25% banana leaves, and 25% grass clippings had significantly higher nitrogen content than treatments with 75 % goat manure + 25% sawdust. This could be due to the lower nitrogen content of sawdust, as reported by Domínguez, Edwards, and Webster (2000).

Guerrero (2010) supports that vermicast has another trace element beyond explainable, making it have an edge compared to other organic fertilizers. Further, he found out that for whatever substrate he fed to earthworm in his studies, its mineral nitrogen content settles down at an average of 1.2 – 2.6%.

Phosphorous Content

Also in table 2 reflects the Phosphorous (K_2O) content of vermi casts. Results showed that Treatments 4 and 3 had significantly higher phosphorous content than Treatments 5, 6, 1, 7 and 2. However, no significant differences were observed between Treatments 4 and 3, between treatments 5, 6, and 1, and between Treatments 7 and 2.

This implies that treatments with 75% goat manure + 25% legume leaves, 25% rice straw grass had significantly higher phosphorous content than the other treatments. In other words, legume leaves and rice straws were noted to have higher Phosphorous content.

Potassium Content

The Potassium content of vermicast is shown in Table 2. Statistical analysis showed significant differences among treatments. Treatments 7, 4, and 5 had significantly higher Potassium content with 1.21%. 1.13% and 1.01% respectively than Treatments 3, 1, 6 and 2 with 0.61%. 0.49%, 0.48% and 0.47% respectively. Results imply that treatments with 75% goat manure + 25% sawdust, 25% legume leaves, and 25% cassava leaves had significantly higher potassium contents than the other treatments. Consistently, the highest Potassium content was observed in Treatment 7, those having a combination of goat manure and sawdust.

Table 2. Nitrogen, Phosphorous, and Potassium Content of Vermicast Using Different Substrates

TREATMENTS	Nitrogen Content (%)	Phosphorous Content (%)	Potassium Content (%)
T1	1.47[a]	1.08[b]	0.49[b]
T2	1.45[a]	0.79[c]	0.47[b]
T3	1.30[ab]	1.49[a]	0.61[b]
T4	1.43[ab]	1.62[a]	1.13[a]
T5	1.33[ab]	1.18[b]	1.01[a]
T6	1.50[a]	1.10[b]	0.48[b]
T7	1.19[b]	0.80[c]	1.21[a]
F-test	*	**	**
CV (%)	9.29	10.81	19.73

Note: Means of the same superscript are not significant at 0.05 level of significance.

CONCLUSIONS

In light of the findings, it could be concluded that Treatment 7, those having 75% goat manure plus 25% of sawdust, could significantly increase the reproduction rate of earthworms.

A combination of goat manure and banana leaves got the highest Nitrogen content but is comparable to grass clippings, rice straw, legume leaves, cassava leaves, and banana bracts. The least performing substrates when it comes to Nitrogen contents were in treatment with sawdust. The highest Phosphorous content was obtained by Treatments with legume leaves but is comparable to rice straw. The least phosphorous content was noted in treatments with grass clippings and sawdust. The highest Potassium content was observed in Treatments with sawdust and cassava leaves.

RECOMMENDATIONS

Farmers may opt to use as substrates for vermicomposting the combination of 75% goat manure + 25% of sawdust,25%cassava leaves,25%grass clippings and25%banana leaves for higher weight and rate of reproduction of earthworm biomass. Farmers also are encouraged to use 75% goat manure + 25% legume leaves for consistently producing higher NPK contents of Vermicasts. Another trial may be conducted to determine the earthworm's reproduction rate by counting the number of biomass (worms) instead of its weight after 45 days of the stocking.

TRANSLATIONAL RESEARCH

The findings of this study could be effectively translated for the use of farmers in the form of leaflets or brochures. The best combination of substrates for vermicomposting will guide farmers on which farm wastes and debris to be used for optimum production of vermi biomass (reproduction rate) and the nutrient contents in the cast and compost in terms of the available and soluble form of nitrogen (N), phosphorous (P_2O5) and potassium (K_2O)

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