

Growth Performance and Serum Chemistry of Earthworm (*Hyperiodrilus euryaulos*) cultured in different Animal Dung Media

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Target Audience: *Fish Farmers, Researchers*

Abstract

The experiment was conducted to determine the growth performance and serum biochemistry of earthworms cultured in different animal dung manures in a Completely Randomized Design experiment that lasted for 10 weeks, using 315 earthworms. Five treatments, T1, T2, T3, T4 and T5 respectively containing garden soil alone (control) and four animals (poultry, rabbit, pig and goat) manure mixed with garden soil in the ratio of 1: 3 were put into rectangular plastic containers (30 x 42.5x27) cm. The containers were perforated at the base to enable excess water to drain out. 1kg of diet each was formulated with maize offal, wheat offal and PKC in a proportion of 0.35kg, 0.30kg and 0.35kg respectively and were mixed thoroughly with each medium. 63 fry earthworms of similar length were introduced into each treatment that was replicated thrice. 1 litre of water was sprinkled to each treatment once every two days. At the end of the experiment, the control group (T1) was observed to have the least values for all the parameters evaluated. The results indicated that the absence of animal dung did not support growth of the earthworms. Poultry dung- cultured earthworms had the highest gain in weight (15.31g) followed by the goat dung-cultured earthworms (13.41g). The animal dungs improved the final average length with the highest value observed in the group cultured in the poultry dung. There were significant differences ($P < 0.05$) in all the serum chemistry parameters evaluated. The control was observed to have 0.53g/dl total protein, 1.80mg/dl creatinine, 10.03mg/dl urea, 13.34mg/dl cholesterol and 6.12g/dl glucose. Group T3 (rabbit dung medium) earthworms had the highest ($P < 0.05$) urea (58.30mg/dl) and cholesterol (106.15g/dl).

Keywords: *Earthworm, animal manure media,*

Description of the Problem

Research findings revealed that diets containing vegetable products as sole sources of protein were deficient in methionine and lysine and so needed to be supplemented with fishmeal or other unconventional but cheaper protein concentrate (1). Research interest has been awakened in the area of alternative unconventional feed resources, which have comparative nutritive value but are cheaper than the conventional feed ingredients.

Although, Nigeria's livestock industries still make use of appreciable quantity of imported protein feedstuffs especially fishmeal, there is not only growing awareness but practice, among animal scientists, of the evaluation of locally available feeding stuffs of animal origin as substitute to imported ingredients. Such alternatives include the use of grasshopper meal, chicken offal meal and maggot meal to mention a few. Earthworm meal is a non-conventional protein

supplement, which can also be sourced locally.

In recent years, considerable attention has been focused on the potential role of intensive earthworm production (vermiculture) in generating earthworm biomass (vermimex) as protein supplement for livestock and fish production. Earthworm production is gaining much interest globally as an effective and environmentally sound method of increasing the rate of decomposition of organic waste and as a potential valuable product used as aqua and livestock feed.

The research on earthworm as animal protein feed ingredient in animal diet is novel, hence there are scarcity of information on its nutrient requirements. Also, earthworm is a fragile animal, which require careful handling during rearing and measurements of the parameters under investigation. With the increased animal production especially poultry, the dungs or faecal droppings will be useful in earthworm rearing and multiplication. This will not only create job opportunity for earthworm growers but will also increase the usefulness of animal dungs apart from using it grow crops as organic manure.

This research work was aimed at evaluating the growth performance and serum chemistry of earthworms raised in four different animal dung substrates, with the broad objectives of evaluating and comparing the earthworm (*Hyperiadrilus euryaulos*) reared in different animal manure.

Materials and Methods

Experimental location

The experiment was carried out in Rubber Plantation of Michael Okpara University of Agriculture Umudike Teaching and Research farm. Mean annual rainfall 2177/year and temperature range 22-36°C. Relative density is about 50-90% it is on elevation of 122m above sea level and latitude location of 5° 29' and 7° 32' (15)

Experimental materials

Animal dungs from four animal species (Goat, Rabbit, Pig, Poultry) were collected in their dry forms, mixed with top soil in a ratio of 2:3 and then put into rectangular plastic container (30 x 42.5x27) cm designated T1, T2, T3 and T4 respectively. Each treatment was replicated three times given a total experimental unit of 12. This container was perforated for draining excess water. 1kg of diet was formulated with maize offal, wheat offal and PKC in a proportion of 0.35kg, 0.30kg and 0.35kg at the ratio of 7:6:7 mixed thoroughly with each medium. The medium was a mixture of 2kg each of the animal manure with 6kg treated garden soil and then mixed thoroughly with 1 litre of water and then introduced into the container. 315 earth worms whose average length and weight were noted were allocated to the treatments with 63 earthworms per treatment, having 3 replicates. The containers were covered with mosquito net to prevent from escape and predators. The containers were kept under the rubber plant tree so as to provide adequate shade. 1 litre of water was sprinkled on each experimental unit every evening to keep the soil moist.

Harvesting of earthworm

Harvesting was done after 10 weeks of introduction of the fry. It was done in the morning by hand picking to ensure effective recovery of the earthworm. Harvested earthworms were thoroughly rinsed in water and kept in a bowl for 30 minutes to evacuate their guts (3). Thereafter data collection was done and the left over earthworm was oven dried and milled.

Experimental data collection

Growth performance

The mean weight gain was determined by the difference in mean weight between final weights of earthworm at the end of the experiment and initial mean weight of earthworm recorded at the beginning of the

experiment. The mean length and diameter were noted at the beginning and end of the experiment using meter rule. In order to avoid movement while measurement was going on, the earthworm to be measured was stretched out on a broom stick and a mark made on the broom stick. Then the broom stick with the mark was placed on the meter rule to read the mark indicating the length of the earthworm. To measure the diameter, a pair of dividers was placed across the width of the earthworm. Then the pair of dividers with the mark was placed on a vernier calipers to get the diameter.

Serum chemistry

Blood samples were collected by grinding 3gm of harvested earthworms from each media using a plastic mortar and pestle and mixed with silver chloride and then put into a centrifuge at 2000 RPM for ten minutes and the supernatant (15ml) was collected and taken to veterinary laboratory to check the serum biochemistry parameters which include; total protein, creatinine, urea, cholesterol, and glucose.

Determination of Total Protein

Total protein was determined by direct Burette method (4) for *in-vitro* determination of total protein in serum as plasma.

Procedure

1ml of Burette reagent was put into a clean and labeled test tube and 0.02ml of serum sample was added to it and mixed. The mixture was allowed to stand for 10 minutes at room temperature. A standard was prepared by adding 1ml of burette reagent into a clean and labeled test tube and 0.02ml of standard was added to it and mixed. The mixture was allowed to stand for 10minutes at room temperature. The absorbance of both the samples and standard were read against the contents of the blank at 540nm using a digital colorimeter and the total protein concentration was obtained.

Determination of Serum Cholesterol

The serum cholesterol was determined by enzymatic colorimetric method (5) for the *in-vitro* determination of cholesterol in serum using a QCA enzymatic cholesterol test kit (Quimica Clinica Aplicada, Spain).

Procedure

1ml of the working reagent was put into clean and labeled test tubes and added 0.01ml of the sample and mixed properly. The standard was also prepared by adding 1ml of the working reagent into clean and labeled test tubes and 0.01ml of the standard was added and mixed properly. The mixtures were allowed to stand for 10 minutes at room temperature. The absorbance of both the samples and standard were read against a reagent blank at 590nm with a digital colorimeter and the cholesterol count of each sample was obtained.

Determination of Serum Urea

The serum urea was determined by the modified Berthelot-secrecy method for the *in-vitro* determination of urea in serum (6), using QCA enzymatic urea test kit.

Procedure

1ml of urease/salicylate solution (reagent A) was put into a clean test tube. 0.01ml serum was added, mixed properly and was allowed to stand for 5minutes at room temperature. After 5 minutes, 1ml of alkaline hypochlorite solution (reagent B) was added and mixed and allowed to stand for 5 minutes at room temperature. The absorbance was read at 600nm against the contents of the blank with digital colorimeter.

Determination of Serum Glucose

The blood glucose level (mg/dl) was determined using the Accu-check active diabetes monitoring Kit based on the glucose oxide method (7). Glucose was determined by the glucose oxidase reaction.

Statistical model and Data analysis

$$Y = \mu + T_1 + e_{ij}$$

Y=Single observation

μ = Overall mean

T_1 = Effect of treatment

e_{ij} = random error assumed to be identically, independently distributed with zero mean and constant variance.

The data obtained was subjected analysis of variance (ANOVA) in a Completely Randomized Design (CRD). Where significant differences were obtained, means were further subjected to Duncan's Multiple Range test (8) as packaged in (9) for windows: version 17 SPSS Inc.

Results and Discussion

Growth Performance

Table 1 shows the growth performance of earthworm cultured in different animal dung media. There were significant differences ($P < 0.05$) in all the parameters except the initial body measurements. The control (T1) was observed to have the least final length, diameter. There was no positive effect on the length of the earthworms cultured in the medium containing no animal dung as can be evidenced in the negative change in length (-1.15cm). Final average length was observed to be 22.00cm, 21.34cm, 17.66cm and 20.02cm

respectively for poultry, rabbit, pig and goat dung media. The highest increase in length (7.67cm) was observed in the rabbit dung-cultured earthworms followed by those cultured in the poultry dung medium (7.50cm). The poultry dung- cultured earthworms had the highest gain in weight (15.82g) with a daily weight gain of 0.22g/d followed by the goat dung-cultured earthworms (13.91g average weight and daily weight gain of 0.19g/d).

The poor performance observed in the control may be attributed to the lack of adequate nutrients resulting from absence of animal dungs. The results indicate that the animal dungs under investigation could be good sources of nutrients relevant in the growth of earthworms (10). The poultry dungs may have contained substantial amount of nitrogen in the form of urea and this may have supported growth. Also, the rabbit dung was observed to support growth due to coprophagy that encourages the rabbit to pass out faeces rich in nitrogenous compounds as reported by (11). The increase in the weight gain observed in all the treatments were in accordance with (12) who reported that in 45-60 days earthworm culture should double in its weight. It also agrees with (4) who observed a 250% Increase in biomass in 2 months in vermiculture.

Table 1: Growth performance characteristics of earthworm cultured in the different dung media

| Growth parameter | T1 | T2 | T3 | T4 | T5 | S.E.M |
|------------------------------|--------------------|--------------------|---------------------|--------------------|---------------------|-------|
| Initial average length (cm) | 14.45 | 14.50 | 14.67 | 14.33 | 14.51 | 0.44 |
| Final Ave. length (cm) | 13.30 ^d | 22.00 ^a | 21.34 ^{ab} | 17.66 ^c | 20.02 ^{bc} | 0.35 |
| Increase in length (cm) | -1.15 ^e | 7.50 ^b | 7.67 ^a | 3.33 ^d | 5.51 ^c | 0.10 |
| Initial Ave. diameter (mm) | 2.02 | 2.44 | 2.40 | 2.54 | 2.45 | 0.48 |
| Final average diameter (mm) | 2.60 | 3.23 | 3.05 | 3.41 | 3.25 | 0.43 |
| Increase in diameter(mm) | 0.58 ^c | 0.79 ^b | 0.65 ^c | 0.89 ^a | 0.80 ^b | 0.03 |
| Initial body weight (g) | 0.53 | 0.51 | 0.48 | 0.50 | 0.50 | 0.08 |
| Ave.Final weight (g) | 5.34 ^d | 15.82 ^a | 12.51 ^b | 10.98 ^c | 13.91 ^b | 0.68 |
| Ave. weight gain (g) | 4.81 ^d | 15.31 ^a | 12.03 ^b | 10.48 ^c | 13.41 ^{ab} | 1.34 |
| Ave. daily weight gain (g/d) | 0.07 ^c | 0.22 ^a | 0.17 ^{ab} | 0.15 ^b | 0.19 ^{ab} | 0.01 |

a, b, c, d means along the same row with different superscripts are significantly different(P<0.05). Ave.-Average. F.C.R.-Feed conversion ratio. S.E.M-Standard Error of the Means.

Blood Chemistry

The blood chemistry of the earthworms cultured in different animal dung media is shown in Table 2. The earthworms reared in the dung-free (control) medium had the least (P<0.05) values of all parameters investigated. It was observed to have 0.53g/dl total protein, 1.80mg/dl creatinine, 10.03mg/dl urea, 13.34mg/dl cholesterol and 6.12g/dl glucose. T3 (rabbit dung medium) had the highest (P<0.05) urea (58.30mg/dl) and cholesterol (106.15g/dl). The low value of total protein observed in the control medium may be attributed to the absence of protein feed ingredient, which may have caused dietary protein deficiency. The varying animal dung media led to a significant (P<0.05) increase in the serum creatinine, urea and cholesterol. The high level of cholesterol in the animal dung-cultured earthworms over the control could be

attributed to the obstruction of the biliary flow as reported by (13). The levels of urea observed in the animal dungs media were higher than that observed in the control and also were higher than the values observed in chicken (14). This may indicate that the proteins in the media (T2, T3, T4 and T5) were poorly utilized. The high cholesterol also observed in the animal dung media over the control may be an indication that the nutrients in the animal dungs were high in fats. The earthworms cultured in the poultry manure had the least glucose.

The results indicated that the various animal dungs supported positive performance of the earthworms with respect to the length, diameter and average weight gain. The different animal dung media appeared to improve the nutrient composition of the culture media as was evidenced in the growth

performance parameters. The poultry dung medium recorded the highest gain in weight of the earthworms. The earthworms cultured in the rabbit dung medium appeared to be affected most in the blood chemistry parameters evaluated. The low value of total

protein observed in the control medium may be attributed to the absence of protein feed ingredient, which may have caused dietary protein deficiency. The animal dung media supported significant ($P<0.05$) increase in the serum creatinine, urea and cholesterol.

Table 2 Serum Biochemistry of earthworm cultured in different animal dung media

| Parameters | T1 | T2 | T3 | T4 | T5 | S.E.M |
|----------------------|--------------------|--------------------|---------------------|--------------------|--------------------|-------|
| Total protein (g/dl) | 0.53 ^d | 1.10 ^c | 2.54 ^a | 2.02 ^b | 2.57 ^a | 0.18 |
| Creatinine (mg/dl) | 1.80 ^d | 5.00 ^b | 7.55 ^a | 3.63 ^c | 1.96 ^d | 0.62 |
| Urea (mg/dl) | 10.03 ^e | 49.62 ^b | 58.30 ^a | 25.75 ^d | 38.29 ^c | 3.69 |
| Cholesterol (mg/dl) | 13.34 ^e | 56.18 ^c | 106.15 ^a | 40.58 ^d | 92.38 ^b | 7.99 |
| Glucose (mg/dl) | 6.12 ^d | 6.05 ^d | 9.67 ^c | 13.28 ^a | 10.92 ^b | 0.79 |

a,b,c,d,e means along rows with different superscripts are significantly difference ($p<0.05$).
S.E.M-Standard Error of Means

Conclusion and Application

1. From the results, earthworms can be successfully cultured using animal dung mixed with garden soil indicating a venture that can enhance the production of animal protein feedstuff for livestock feed with a concomitant significant reduction in the cost of livestock production in Nigeria.
2. The massive production of earthworms could be a leverage to fish farmers who would find it as ready feed for fishes in the pond, fed raw or dried. Thus reducing overdependence on the exotic commercially formulated fish feed whose exorbitant price cut deep into the farmers' profit.
3. Vermiculture is a potential enterprise that can contribute immensely to the increase in animal protein consumption in Nigeria.

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