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Bondhare SO
Assistant Professor, Department of Zoology, Hutatma Jayvantrao Patil Mahavidyalaya, Himaytnagar, Nanded, Maharashtra, India

Desai RB
Head, Department of Zoology, Mahatma Gandhi Vidyalaya, Ahmedpur, Latur, Maharashtra, India

Correspondence
Desai RB
Head, Department of Zoology, Mahatma Gandhi Vidyalaya, Ahmedpur, Latur, Maharashtra, India

Comparision of life cycle of earthworms *Eisenia fetida* and *Lumbricus rubellus* under controlled condition, in Nanded district

Bondhare SO and Desai RB

Abstract
The study was initiated and reproductive efficiency of vermicomposting worms to see the growth, reproductive biology and life cycle of the vermicomposting earthworms (*Eisenia fetida* and *Lumbricus rubellus*) at the lab Department of Chemistry during 25 Jan to 20 May 2017. Determination of cocoon production rate, hatching success, number of hatchling and growth of *Eisenia fetida* and *Lumbricus rubellus* in the Cow dung media was conducted with plastic containers. The experiment was designed under the single treatment Cow dung with three replications. Clitellum development started earlier in 3rd week of *Eisenia fetida* than *Lumbricus rubellus* (4th week). Number of cocoons produced per earthworm (21.8±3.0), mean number of hatchlings from one cocoon (3.3±0.57), hatching success rate (86.6%) was recorded higher of *Eisenia fetida.*
Cocoon production of E. *fetida* was started to release cocoon on the 6th week and *Lumbricus rubellus* was started to release cocoon on the 7th week. Cocoon production ceased after 12 week for *Eisenia fetida* and 9 week *Lumbricus rubellus.* Maximum weights achieved per earthworm (1116±16.4 mg), net weight gain per earthworm (819±12 mg), growth rate per worm per day (17.43±1.4 mg) were higher in *Lumbricus rubellus* comparison to *Eisenia fetida.* The overall result of this study showed that better hatching success of cocoons and number of hatchling per cocoon were recorded by *Eisenia fetida* earthworm species.

Keywords: *Eisenia fetida, Lumbricus rubellus,* earthworms, life cycle

Introduction
The term Earthworm refers to a specific group of invertebrates within the taxonomic phylum Annelida. Earthworms belong to the Oligochaetes-which means 'few bristles' and are represented by 27 species of earthworm here in the UK. Worldwide there are over 3,000 species described- and advances in taxonomy using DNA is changing this, highlighting new species and new subspecies. Earthworms are, as their name suggests, terrestrial, their skin is permeable and they need a moist environment so they don't dry out. The different species of earthworms have individual requirements just as a dog has from a cat, some earthworm species live in compost, some live in permanent burrows deep deep down in the soil, others are content with the middle ground and make complex networks of tunnels as they explore the earth.
An earthworm is a tube-shaped, segmented worm found in the phylum Annelida. They have a world-wide distribution and are commonly found living in soil, feeding on live and dead organic matter. An earthworm's digestive system runs through the length of its body. It conducts respiration through its skin. It has a double transport system composed of coelomic fluid that moves within the fluid-filled coelom and a simple, closed blood circulatory system. Earthworms are macroscopic clitellate oligochaete annelids that live in soil. They are segmented worms, bilaterally symmetrical, with an external gland (clitellum) for producing the egg case (cocoon), a sensory lobe in front of the mouth (prostomium), and an anus at the end of the animal body, with a small number of bristles (setae) on each segment. They are hermaphrodite animals, and reproduction normally occurs through copulation and cross-fertilization, following which each of the mated individuals produces cocoons containing 1-20 fertilized ova. The resistant cocoons, which are tiny and roughly lemon-shaped, with shape differing between species, are usually deposited near the soil surface, except in dry weather when they are laid at deeper layers. Cocoons hatch after an incubation period that varies according to the earthworm species and environmental conditions.

Hatchling earthworms, unpigmented and only a few millimeters in length on emerging from the cocoons, gain their adult pigmentation within a few days. Assuming favorable conditions, they reach sexual maturity within several weeks after emergence.

Mature individuals of most vermicomposting species can be distinguished easily by the presence of the clitellum, the pale or dark colored swollen band located behind the genital pores. The clitellum secretes the fibrous cocoon, and the clitellar gland cells produce a nutritive albuminous fluid that fills the cocoon. Earthworms display indeterminate growth and can continue to grow in size after completing their sexual development although they do not add segments. According to Reynolds and Wetzel [1], there are more than 8300 species in the Oligochaeta, of which about half are terrestrial earthworms. On the basis of their feeding and burrowing strategies, into three ecological categories: epigeic, anecic, and endogeic [2]. Endogeic (soil feeders) and anecic species (burrowers) live in the soil and consume a mixture of soil and organic matter, and thus excrete organomineral feces. Epigeic species of earthworms are litter dwellers and litter transformers; they live in organic soil horizons, in or near the surface litter, and feed primarily on coarse particulate organic matter.

They ingest large amounts of under composed litter and excrete holorganic fecal pellets. These species are small in body size and uniformly pigmented with high metabolic and reproductive rates, which represent adaptations to the highly variable environmental conditions at the soil surface. Different species of earthworms have different life histories, occupy different ecological niches. Loh *et al*. [3] reported higher cocoon production and weight gain by *E. fetida* in cattle waste than in goat waste. Gunadi *et al*. [4] have studied growth, reproduction, and mortality of *E. foetida* for over a year in solid manure, pig manure, and supermarket waste solids. Worms could not survive in fresh cattle solids, pig solids, fruit wastes, or vegetable wastes. The growth of E. foetida in pig wastes was faster than in cattle solids. Understanding the growth and reproductive efficiency of vermicomposting worms in various substrates is highly essential for effective utilization of earthworms in sustainable waste management system [5-6].

As huge amount of wastes can be managed through more population of earthworms [7] reproductive and growth performance of various species of earthworms in a range of substrates can act as useful biomarkers to measure the efficiency of an earthworm species in vermicomposting or earthworm based biotechnology [8]. Neuhauser *et al*. [9] have reported that the weight gain by *Eisenia fetida* is positively correlated with food type. Similarly, Nath *et al*. [10, 11] have also described substrates that provide earthworms with sufficient amount of easily metabolizable organic matter, facilitate growth and reproduction. Hence, in the present study, to see the growth, reproductive biology and life cycle of the vermicomposting earthworms.

Materials and Methods

Life cycle experiments were carried out in the lab Department of Chemistry with average room temperature of 21-28°C and 60 to 80% moisture of the culture media during 25 Jan to 20 May 2017. For moisture determination, the sample substrates were taken regularly (at 3 days interval) weighed, oven dried at 105°C and cooled in desiccators for 1 hr and reweighed. The difference between moist and dried samples were taken and then the moisture content of beddings was adjusted to 60 to 80% and the temperature was kept within the range of 21-28°C throughout the study period [12]. In this experiment was designated as single treatment and two earthworm species *(Eisenia fetida* and *Lumbricus rubellus)* with three replications. For experimentation 500g Cow dung was filled in the container. Twenty each earthworm species having individual live weight (225 mg of *Eisenia fetida* and 284 mg of *Lumbricus rubellus)* were released into each experimental container.

Biomass, Maturation and Cocoon Production Date

Five hatchlings of earthworm species in good health condition were taken from the above containers for reproductive potential determination. The hatchlings were rinsed with distilled water to remove any adhering material, dried briefly on paper towel, weighed on electronic balance and finally introduced in each respective experimental container. Three replicates were established. To monitor the growth and maturation progress weekly, the biomass was measured in batch of earthworm in each container and three phases of the life cycle were observed, Pre-clitelleate (incipient development of the clitellum, identified by appearance of tuber culapubertasis), clitelleate (well-developed clitellum) and regression (loss of the clitellum). At the same time, the first cocoon production date for each worm was also determined. No additional feed was added at any stage during the study period. On the basis of the obtained data on the biomass of the worms other parameters of earthworm such as growth rate or biomass increase rate (g/ earthworm/day), maximum weight achieved, and net weight gain were calculated.

Reproductive Potential (Cocoon Production Rate) Determination

The experiments were conducted in cylindrical plastic containers. Each container was filled with waste material based on the feeding rate explanation of [13]. Three replicates were prepared for each substrate and worm combination. Two freshly clitellated species, in good health condition, were collected from the stock culture and rinsed with distilled water to remove any adhering material, dried briefly on paper towel, weighed with electronic balance and finally introduced in each container containing. The substrate in treatment container was examined daily in order to determine the onset of cocoon production. Once the cocoons appeared, they were separated by hand sorting, washed lightly in distilled water and counted so as to determine total number of cocoon and the fecundity or reproductive rate (cocoon/worm/day).

Cocoon Incubation Period, Number of Hatchlings and Hatching Success

To determine the incubation period (time lapse from cocoon formation until the first hatchling emerged in days), hatching success (total number of hatched cocoons), and number of hatchlings per cocoons, fifteen freshly laid cocoons from treatment were taken from the above containers and placed in containers which contained the same material in which their parents were reared. Three replicates were prepared. The beddings were observed daily for the emergence of hatchlings in order to determine the incubation period. As soon as the appearance of hatchling started, they were removed daily using a fine painting brush and counted by hand sorting in order to determine the total number of hatchlings that emerged from a single cocoon. The number of unhatched cocoons was also counted in order to determine the hatching success of cocoons.

Results and Discussion

Growth Rate of Earthworm Species

The growth rate of two earthworm species in investigating during the study period is given in Table 1. The biomass of earthworm species showed progressive raise up to 6th week in E. *fetida* and 7th week in *Lumbricus rubellus*. In E. *fetida* where maximum growth attained, E. *fetida* achieved their maximum weight achieved 889±90 mg/worm at a growth rate of 16.3±0.52 mg/worm/day in the 6th week. In *Lumbricus rubellus* where maximum growth attained, *Lumbricus rubellus* achieved their maximum weight achieved 1116±16.4 mg/ worm at a growth rate of 17.43±1.4 mg/worm/day in the 7th week. Regarding the sexual maturity (clitellum development), E. *fetida* was preclitelated on the second week and mature individual with clitellum totally developed started to emerge on the 3rd week and *Lumbricus rubellus* was developed in 4th week of this experiment.

Table 1: Growth parameters of two earthworm species.

Earthworm species	Initial weight/ earthworm (mg)	Maximum weight achieved/ earthworm (mg)	Maximum weight achieved on	Net weight gain/ earthworm (mg)	Growth rate/ worm/day (mg)	Clitellum development started in
Eisenia fetida	196±69	889±90	6th week	686±22	16.3±0.52	3 rd week
Lumbricus rubellus	297±4.1	1116±16.4	7th week	819±12	17.43±1.4	4th week

Note: All values were introduced as the mean ± SD (standard deviation).

Edwards *et al*. [14] and Monroy *et al*. [15] also reported that rapid pre-reproductive phase of growth, followed by a phase of progressive biomass and growth reduction once sexual maturity was attained. These losses in worm biomass might be associated with the exhaustion of food. As supported by [9] who reported that when E. *fetida* received food below a maintenance level, it lost weight. The weight reduction was also occurred because the earthworms attained the matured stage. So, they utilized the energy for reproduction purpose such as laying eggs, mating and cocoon formation [6]. Reinecke *et al*. [16] reported a maximum weight gain for *Lumbricus rubellus* of 150 mg per week, at 25°C. Hait and Tare [17] reported that temperature influences the biology of the earthworm by modifying metabolic activity.

Table 2: Rate of cocoon production by two earthworm species.

Earthworm species	Cocoon production started in	Total no. of cocoons produced	No. of cocoons produced/ earthworm	No. of cocoons produced/ earthworm/day	Cocoon production ceased after
Eisenia fetida	6th week	109±14.9 (12 weeks)	21.8±3.0	0.39±0.05	12 th week
Lumbricus rubellus	7th week	1016±31 (9 weeks)	12.61±1.0	0.21 ± 0.07	9th week

Note: All values were introduced as the mean ± SD (standard deviation).

Rate of Cocoon Production by Earthworm Species

In this experiment, E. *fetida* was started to release cocoon on the 4th week and *Lumbricus rubellus* was started to release cocoon on the 5th week (Table 2). The highest total number of cocoon was attained in *Lumbricus rubellus* (1016±31 cocoons at a reproduction rate of 0.21 ± 0.07 cocoon/earthworm/day) at 9 weeks and E. *fetida* (109 ± 14.9 cocoons at cocoon production rate of 0.39±0.05 cocoon/earthworm/day) was recorded in this study. Cocoon production ceased after 12th week of *Eisenia fetida* and *Lumbricus rubellus* cocoon production was ceased in 9th week. Giraddi *et al*. [18] who reported that better hatching success of cocoons that indicate the cocoon viability. *Lumbricus rubellus* reached sexual maturity in as little as five weeks compared with E. *fetida* which took 6-8 weeks to produce its first cocoon [19]. Reinecke *et al*. [16] who reported that *Lumbricus rubellus* started producing cocoons after 46 days. This is a shorter time than the 6-8 weeks previously quoted for E. *fetida* [20], and the 51 days reported for *Lumbricus rubellus* [21].

Incubation Period and Hatching Performance of Cocoons

The incubation period range of two earthworm species were completed in 18-26 days of *Eisenia fetida* and 12-21 days of *Lumbricus rubellus.* The mean number of hatchlings from one cocoon was 3.3±0.57 in *Eisenia fetida* which comparison 2.23 ± 0.18 in *Lumbricus rubellus.* The cocoons produced generally demonstrated good hatching performance in *Eisenia fetida* (86.6%) than *Lumbricus rubellus* (75.3%) (Table 3). Shalabi [22] who reported that the average standard time taken to reach sexual maturity for E. *fetida* was about 70 days. The result also varied from the result of Venter *et al*. [23] who recorded 60 days for E. *fetida* to reach sexual maturity.

Table 3: Incubation period and hatching performance of cocoons by two earthworm species.

Earthworm	Incubation period (in days)	Number of hatchlings from one cocoon	Mean number of hatchlings from one cocoon	Hatching success
Eisenia fetida	18-26	2-4	3.3±0.57	86.6%
Lumbricus rubellus	12-21	2-3	2.23 ± 0.18	75.3%

Note: All values were introduced as the mean ± SD (standard deviation).

Conclusion

The major benefits of earthworm activities to soil fertility for agriculture. The reproductive ability of *Eisenia fetida* is higher than *Lumbricus rubellus* it. However, to get accurate knowledge of the full potential of the compost worm as waste processor, it is essential that the life cycle of the worm on waste be studied thoroughly. *Eisenia fetida* is very suitable as waste decomposer.

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