Pergamon

PII: S0038-0717(96)00276-3

Soil Biol. Biochem. Vol. 29, No. 3/4, pp. 743–746, 1997

Printed in Great Britain
0038-0717/97 $17.00 + 0.00

EFFECTS OF STOCKING RATE AND MOISTURE CONTENT ON THE GROWTH AND MATURATION OF *EISENIA ANDREI* (OLIGOCHAETA) IN PIG MANURE

J. DOMÍNGUEZ[1]* and C. A. EDWARDS[2]

[1]Dpto. Ecología y Biología Animal, Universidade de Vigo, Apt. 874, E-36200 Vigo, Spain and
[2]Dpt. Entomology, Ohio State University, 103 Botany and Zoology Building, 1735 Neil Av., Columbus, OH 43210-1220, U.S.A.

(Accepted 23 September 1996)

Summary—The effect of stocking rate and moisture content on the growth and maturation of *Eisenia andrei* (Bouché, 1972) in pig manure was studied in laboratory trials at 20°C. Six moisture contents (65, 70, 75, 80, 85 or 90%) and five stocking rates (1, 2, 4, 8 or 16 worms, each per the same volume of substrate) were tested. Growth and maturation of earthworms were monitored over 44 days. The 85% moisture content was found to be most favourable for earthworm growth. The 80 and 90% moisture contents were found to be adequate. Eight earthworms per 43.61 g (dry wt) was found to be the most favourable inoculation density. © 1997 Elsevier Science Ltd

INTRODUCTION

The very large amounts of organic waste that are produced by intensive animal production cause major disposal problems and a considerable potential for pollution of surface and groundwaters. The potential of using earthworms in the digestion of raw organic wastes has been established in small and large-scale projects by many researchers (e.g. Mitchell *et al.*, 1977; Hartenstein *et al.*, 1979; Neuhauser *et al.*, 1980; Kaplan *et al.*, 1980; Edwards, 1988). Environmental conditions and earthworm population density are known to affect earthworm growth and reproduction. Although moisture requirements and moisture preferences of *Eisenia fetida* are considered to be well known (Kaplan *et al.*, 1980; Reinecke and Venter, 1987; Edwards, 1988) the physicochemical properties of different organic wastes indicate that these preferences can vary among substrates. In addition, the polemic taxonomic situation between *Eisenia fetida* and *Eisenia andrei* was another reason to make this study.

For the same reasons it is important to determine the optimum worm density for *Eisenia andrei* in pig manure. Although there are some studies about this topic, the substrates are generally cow or horse manure (Neuhauser *et al.*, 1980; Edwards, 1988; Reinecke and Viljoen, 1990).

The present study was undertaken to determine the effect of worm density and moisture content on the growth and maturation of *Eisenia andrei* (Bouché, 1972) in pig manure in laboratory trials at 20°C.

*Author for correspondence: 36200-VIGO-(Spain), Fax: +34-86-812556, e-mail: jdguez@setei.uvigo.es

MATERIALS AND METHODS

Juvenile specimens of *Eisenia andrei*, weighing less than 100 mg live wt, were obtained from a stock culture. Four individuals were placed in 250 ml plastic dishes and fed 100 g of a mixture of pig manure (85 g) and maple leaves (15 g). Four replicates for each moisture content were used to determine the moisture requirements of *Eisenia andrei* in pig manure. 100 g of the mixture at 60% moisture content were placed into each dish. Water was added to the medium in the different dishes to obtain the following moisture contents: 65, 70, 75, 80, 85 or 90%. The worms were fed no additional substrate for the duration of the experiment (44 days). The survival and biomass of the worms and the presence or absence of clitellum at different moisture contents were monitored every 4 days. The moisture content of the substrate was determined every 2 days using an infrared moisture detector and held constant by adding water.

In a second experiment, conducted under similar conditions, with the same pig solids and leaf substrate, the effect of worm density was studied. Five different stocking rates (1, 2, 4, 8 or 16 worms each) were placed in 250 ml plastic dishes and fed 150 g of substrate, at 80% moisture content. Four replicates were set up at each density. Every 4 days the worms were weighed and their maturity state was determined over 48 days.

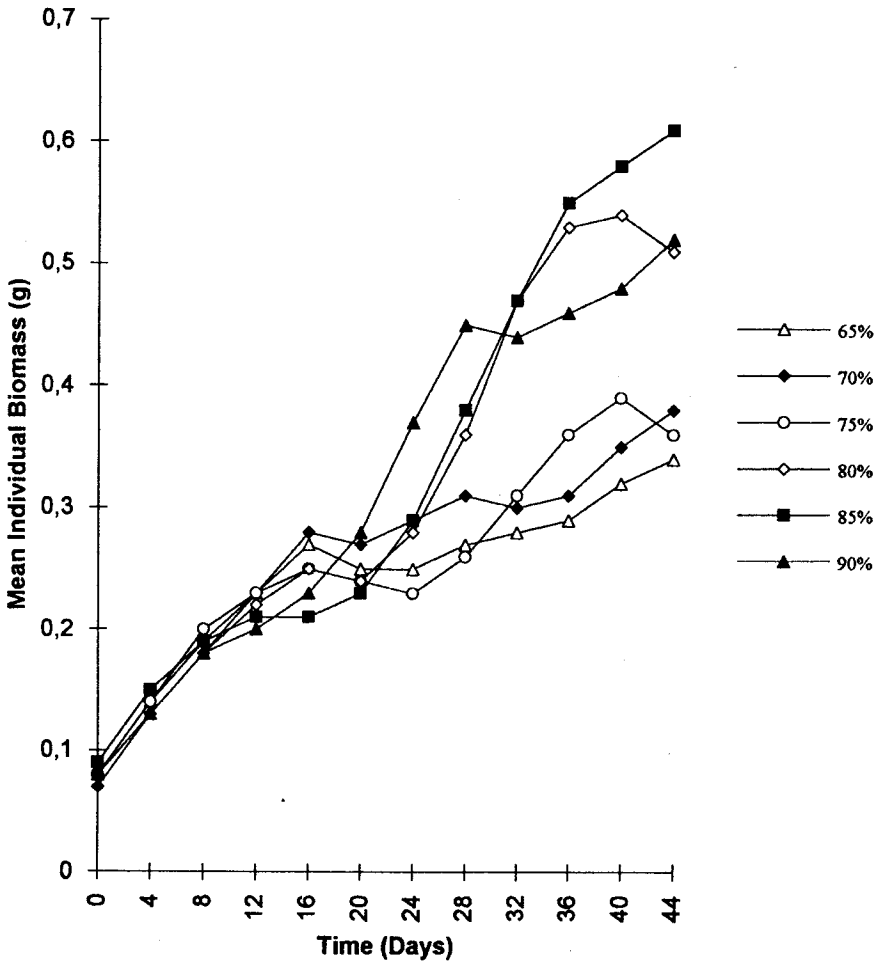

Fig. 1. Growth curves of *E. andrei* on a pig manure mixture at different moisture contents over 44 days at 20°C.

RESULTS

Influence of moisture

Figure 1 summarizes the mean individual growth of *E. andrei* in pig manure at 65, 70, 75, 80, 85 and 90% moisture contents. Growth at the six moisture contents was similar initially until day 16. At the end of the experiment, growth at the three upper moisture contents was higher ($P \leq 0.01$) and the maximum growth ocurred at 85% moisture ($P \leq 0.05$). Table 1 shows the relationship between moisture contents and two growth data: the maximum weight achieved and the growth rate (mg wt gain worm^{-1} d^{-1}). Until 85% moisture the higher moisture conditions clearly facilitated growth, as measured by the increase in biomass. Increased moisture up to 90% clearly accelerated the development of sexual maturity, whereas not all the worms at 65–75% developed a clitellum even after 44 days. Additionally, earthworms at sexual maturity had greater biomass at higher moisture contents compared to worms grown at lower moisture contents.

Stocking rate

The influence of five stocking rates in the same unit of waste on the individual growth of worms is summarized in Fig. 2. Whereas individual worms grew more and faster at the lowest population densities, the total biomass production was maximum

Table 1. Growth and maturation of *Eisenia andrei* in pig manure at different moisture contents

Moisture content (%)	Growth rate (mg biomass d^{-1})	Max. indiv. biomass (mg) ± SD	Mean maturation size (mg) ±SD
65	5.80	335 ± 72 a[1]	268 ± 12 a
70	7.02	379 ± 90 a	295 ± 15 a
75	7.77	391 ± 90 a	304 ± 12 a
80	11.47	537 ± 162 b	467 ± 36 b
85	11.81	608 ± 181 c*	468 ± 31 b
90	9.93	515 ± 149 b	449 ± 46 b

[1]Means in the same column followed by different letters are significantly different at $P \leq 0.01$ (ANOVA and Fisher LSD multiple comparison test).
* $P \leq 0.05$.

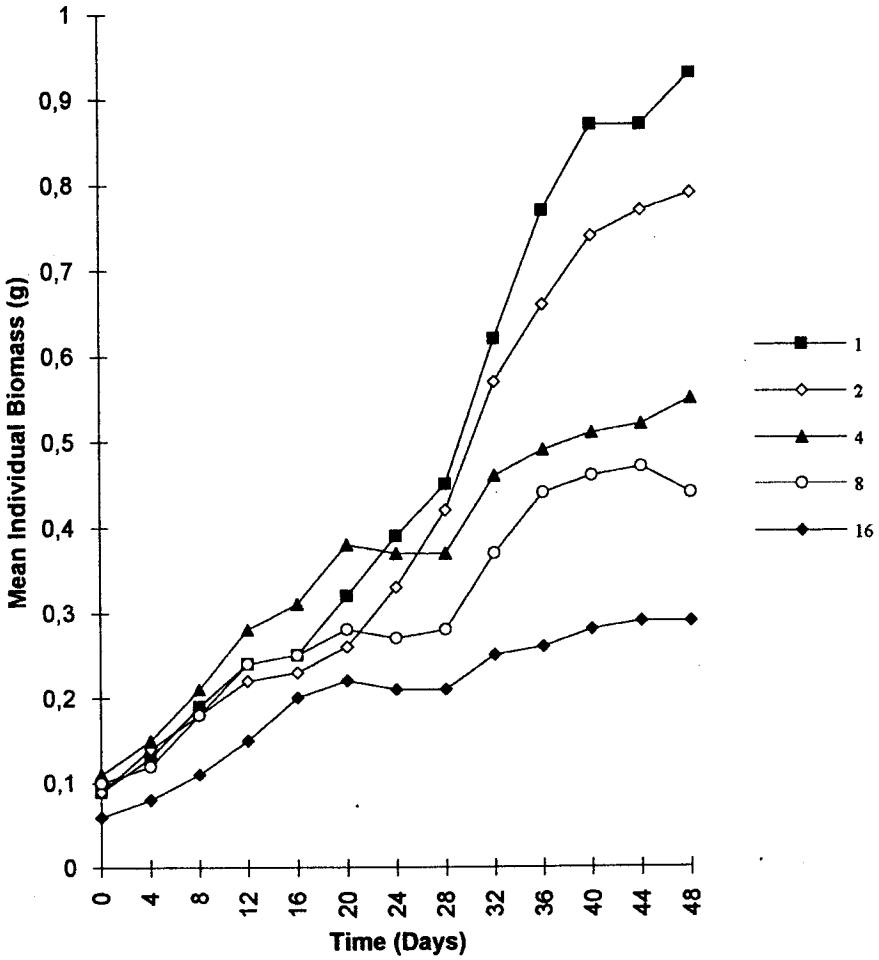

Fig. 2. Mean individual growth of *E. andrei* in a pig manure mixture at five different stocking rates (1, 2, 4, 8 or 16 earthworms).

at the greatest population density tested (16 worms). Clearly high maturation size is related to high growth rate (Table 2). The worms became sexually mature at a younger age at the higher stocking rates, although not all of the worms had developed a clitellum after 48 days. At lower stocking rates all the worms became sexually mature, although the time to reach sexual maturity was longer. With a stocking rate of four worms, all of the worms became sexually mature at day 36, and 40% of worms became sexually mature at day 24.

DISCUSSION

Influence of moisture

We show that *E. andrei* cultured in pig manure grew and matured between 65 and 90% moisture content, the optimum being 85%. There is a direct relationship between the moisture content and the growth rate of earthworms. According to Reinecke and Venter (1985), it is axiomatic that a lowering of the growth rate due to low moisture conditions would also retard sexual development. Worms of

Table 2. Growth and maturation of *Eisenia andrei* in pig manure at five different stocking rates

Stocking rate	Growth rate (mg biomass d^{-1})	Max. indiv. biomass (mg) ± SD	Mean maturation size (mg) ± SD
1	17.54	930 ± 84a[1]	772.5 ± 40 a
2	14.52	791 ± 71 b	662.5 ± 66 a
4	9.10	546 ± 39 c	366.7 ± 10 b
8	8.43	468 ± 35 c	265.0 ± 15 b
16	5.11	288 ± 21 d	284.2 ± 23 b

[1] Means in the same column followed by different letters are significantly different at $P \leq 0.01$ (ANOVA and Fisher LSD multiple comparison test).

the same age developed clitella at different ages in the different moisture conditions.

The temperature and moisture content of the waste are the two most important environmental factors in vermicomposting processes. Dresser and McKee (1980) found that moisture contents between 50 and 80% are the most appropriate for vermicomposting processes. *E. fetida* can survive in a moisture range between 50 and 90% (Edwards *et al.*, 1985; Sims and Gerard, 1985) and grows more rapidly between 80 and 90% in animal wastes (Edwards *et al.*, 1985). Kaplan (1980) achieved a greater worm biomass when reared in sewage sludge with a moisture content between 70 and 85%. Loehr *et al.* (1985) recorded a slightly higher optimum moisture contents in sewage sludge (85 and 90%). Reinecke and Venter (1985) concluded that the optimum moisture content for *E. fetida* lies well above 70% in cow manure.

Influence of stocking rate

Even when the physical conditions are ideal for vermicomposting, problems can develop due to overcrowding. This study clearly showed that, when *E. andrei* was grown at different population densities, the worms in the crowded dishes grew more slowly and with a lower final bodyweight, although the total weight of worm biomass produced per unit of waste was greater (Fig. 2). Our study agrees with the results of Reinecke and Viljoen (1990). Maturation rate was also affected by stocking rate. Worms of the same age developed clitella at different times in the different stocking rates.

REFERENCES

Dresser C. and McKee I. (1980) Compendium on solid waste management by vermicomposting. Prepared for the Municipal Environmental Research Laboratory, Office of Research and Development, Environmental Protection Agency, Cincinnati.

Edwards C. A., Burrows I., Fletcher K. E. and Jones B. A. (1985) The use of earthworms for composting farm wastes. In *Composting of Agricultural and Other Wastes* (J. K. R. Grasser, Eds), pp. 229–241. Elsevier Applied Science, Oxford.

Edwards C. A. (1988) Breakdown of animal, vegetable and industrial organic wastes by earthworms. In *Earthworms in Waste and Environmental Management* (C. A. Edwards and E. F. Neuhauser, Eds), pp. 21–31. SPB, The Hague.

Hartenstein R., Neuhauser E. F. and Kaplan D. L. (1979) Reproductive potential of the earthworm *Eisenia foetida*. *Oecologia* **43**, 329–340.

Kaplan D. L., Hartenstein R., Neuhauser E. F. and Malecki M. R. (1980) Physicochemical requirements in the environment of the earthworm *Eisenia foetida*. *Soil Biology and Biochemistry* **12**, 347–352.

Loehr R. C., Neuhauser E. F. and Malecki R. (1985) Factors affecting the vermistabilization process. Temperature, moisture content and polyculture. *Water Research and Technology* **19**, 1311–1317.

Mitchell M. J., Mulligan R. M., Hartenstein R. and Neuhauser E. F. (1977) Conversion of sludges into "topsoils" by earthworms. *Compost Science* **18**(4), 17–26.

Neuhauser E. F., Hartenstein R. and Kaplan D. L. (1980) Growth of the earthworm *Eisenia foetida* in relation to population density and food rationing. *Oikos* **35**, 93–98.

Reinecke A. J. and Venter J. M. (1985) The influence of moisture on the growth and reproduction of the compost worm *Eisenia fetida* (Oligochaeta). *Revue de Écologie et Biologie du Sol* **22**, 473–481.

Reinecke A. J. and Venter J. M. (1987) Moisture preferences, growth and reproduction of the compost worm *Eisenia fetida* (Oligochaeta). *Biology and Fertility of Soils* **3**, 135–141.

Reinecke A. J. and Viljoen S. A. (1990) The influence of worm density on growth and cocoon production of the compost worm *Eisenia fetida* (Oligochaeta). *Revue de Écologie et Biologie du Sol* **27**, 221–230.

Sims R. W. and Gerard B. M. (1985) Earthworms. In *Synopses of the British Fauna (New Series)* **31** (D. M. Kermack and R. S. K. Barnes, Eds), 171 pp. Natural History Museum, London.