

# Gut load and food-retention time in the earthworms Lumbricus festivus and L. castaneus: A field study

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Received July 27, 1990

Summary. Lumbricus festivus and L. castaneus consume dung. In the field, below cow pats, their gut loads were about 0.15 and 0.14 g dry weight  $g^{-1}$  ash-free dry weight of worm, respectively, but in "free" soil the loads were higher, about 0.21 and 0.19 g  $g^{-1}$  ash-free dry weight of worm. The gut contents of dung were lighter than the total ingested material, at about 0.10 and 0.07 g dry weight  $g^{-1}$  ash-free dry weight of worm, respectively. Field experiments showed that the retention time of dung ranged from >9 to 15 h for *L*. *festivus*, and from >3 to 6 h for L. castaneus. The experiments also indicated that L. festivus exploited 20- and 36-day-old dung in different ways, since the gut load was lower in those worms consuming 20-day-old dung than in those consuming 36- to 40-day-old dung. On the basis of these results the calculated consumption rate for L. festivus is 0.08 g dung  $day^{-1}g$  live weight of worm<sup>-1</sup>, and for L. castaneus 0.15 g dung day<sup>-1</sup>, with retention times assumed to be at maximum, 15 h, for L. festivus and 6 h for L. castaneus. These calculations indicate that our field population of worms  $(75 \text{ g m}^{-2})$  consumes 10-15 t dung ha<sup>-1</sup> 180 days<sup>-1</sup>, corresponding to the amount of dung produced by 2-3 dairy cows.

**Key words:** Gut load – Retention time – Dung – Earthworms – Lumbricus festivus – Lumbricus castaneus

Earthworms play an important role in many temperate ecosystems. Guild (1955) reported that earthworms in pastures may ingest 22-27 t ha<sup>-1</sup> year<sup>-1</sup> of cow dung, and Satchell (1967) calculated that earthworms in a deciduous forest may consume an annual leaf-fall of 3 t ha<sup>-1</sup> in approximately 3 months. The estimates were based on field population studies and consumption rates measured in the laboratory.

The objectives of the present study were to measure the gut load and food-retention time in the earthworms *L. festivus* and *L. castaneus* in the field, and to calculate the consumption rate. The two species are common surface-feeding detritivores in Danish pastures. Cattle dung was offered as food in this study, since (1) earthworms are known to ingest dung (Barley 1959; Holter and Hendriksen 1988; Hendriksen 1991); (2) they aggregate below cow pats (Holter 1983); (3) they play an important role in the disappearance of dung pats (Holter and Hendriksen 1988); and (4) dung lends itself to marking by chromic oxide (Holter and Hendriksen 1987), thus allowing a study of ingestion rates.

## Material and methods

The study site is a permanent pasture at Strødam about 35 km north of Copenhagen, Denmark. The soil is a dark, poorly drained, fen soil, with a pH of about 6.0, and loss on ignition of 33.2%.

Dung consumption by the earthworms was studied by marking the dung with chromic oxide, a water-insoluble and biologically inert marker, which has been used repeatedly in feeding studies of mammals (Van Soest 1982) and invertebrates (McGinnis and Kasting 1964a, b; Holter 1973), and in studying the disapperance of organic matter from cattler dung pats (Holter and Hendriksen 1987). The amount of dung ingested by individual earthworms was calculated from their content of chromic oxide, analysed according to McGinnis and Kasting (1964b). Earthworm sizes were measured as ash-free dry weight. In all studies, artificially pats were made of 2 kg fresh dung, obtained from a cowhouse at Trollesminde, and 10 g chromic oxide was added before homogenization in a Kenwood kitchen machine. Pats 21 cm in diameter were placed in the field, resting on nylon nets (mesh size 7 mm).

To study the total gut load (dung and other material ingested), worms were dug out from below 36- or 40-day-old pats. These specimens were allowed to void their gut contents into clean Petri dishes for 3 days at  $10^{\circ}$ C. The collected faeces were dried at  $60^{\circ}$ C and weighed, and the ash contents were determined after 2 h at  $500^{\circ}$ C. Additional worms were collected from soil without dung pats. To measure the gut load of dung, worms from below the marked pats were dropped into boiling water and the amount of ingested dung were determined. Other worms from below the marked pats were allowed to void their gut contents as previously described, and their weight, and ash and dung contents were determined.

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The retention time of dung was studied in two experiments. (1) Two batches of dung pats, 16 marked and 16 unmarked, were placed in the field, allowing about 200 m between batches. After 20 or 36 days, eight of the marked pats were lifted and carried on their nets to the site of the unmarked pats, which had been removed, and the marked pats were placed exactly in the vacated positions. After 3, 6, 9, 12, 15, 18, 21, and 24 h, one of these pats was removed, and the worms below were collected, killed in boiling water, and kept in a deep freezer until analysed for dung content. The mean daily temperature during the 20-day period varied between 7.3 and 12.9 °C (mean 11.2 °C), and during the 36-day period between 10.1 °C and 14.4 °C (mean 12.4 °C). (2) The other experiment was carried out in eight wooden boxes ( $0.5 \times 0.5 \times 0.3$  m) containing a 10-cm layer of sward and topsoil from the pasture, which were placed outdoors near the Copenhagen laboratory. Eight marked pats were placed in the field and eight identical but unmarked pats were placed in the boxes. After 20 days, 10-15 L. festivus, captured in Bøgemosen, were added to each box. After 40 days the pats in the boxes were replaced with the marked ones. After 3, 6, 9, 12, 15, 18, 21, and 24 h one pat was removed from each box, the worms in the box collected, and the dung contents determined as above. The mean daily temperature varied between 9.8 °C and 17.8 °C (mean 15.7 °C).

#### Results

Table 1 shows the total gut loads present. Thsoe from below the cow pats, both *L. festivus* and *L. castaneus*, were significantly lower than those of worms collected from the free soil (P < 0.01, *t*-test). Similarly, the ash contents of faeces from *L. festivus* were significantly higher in worms from the free soil than in those from beneath pats (P < 0.001). The gut contents of dung were  $0.100 \pm 0.01$ and  $0.07 \pm 0.04$  g g<sup>-1</sup> ash-free dry weight of worm for *L. festivus* and *L. castaneus*, respectively. For both species this was significantly lower than the gut load of total ingested material (P < 0.001). The relative gut loads did not differ between the two species, although there was more material in the gut of adult *L. festivus* than in adult *L. castaneus*, since *L. festivus* was the larger worm.

Figures 1 and 2 show the amounts of marked dung in *L. festivus* and *L. castaneus* after different periods of exposure to the dung. An analysis of the ranked data based on general linear models (Zar 1984) showed that *L. festivus* exploited 20- and 36-day-old dung in different ways, since the two groups of data were significantly different (P < 0.05), while there seemed to be no differences between the two experiments. In contrast, *L castaneus* appeared to exploit 20- and 36-day-old dung in the same way, since no significant difference was found. Kruskal-Wallis tests applied to the results for *L. festivus* showed

**Table 1.** Gut loads (g dry weight  $g^{-1}$  ash-free dry weight) of *Lumbricus festivus* and *L. castaneus* collected below dung pats or in soil

	Gut load	Ash content (% dry weight)
L. festivus		
Below pats	$0.15 \pm 0.01$	$28.3 \pm 2.5$
Free soil	$\textbf{0.21} \pm \textbf{0.03}$	$49.6 \pm 2.3$
L. castaneus		
Below pats	$0.14 \pm 0.06$	_
Free soil	$0.19 \pm 0.03$	_

no significant differences between the amounts of 20-day-old dung (P > 0.1), but there was a difference for the older dung (data from two experiments pooled). A non-parametric multiple comparison procedure (Dunn 1964) showed that this difference was the result of significant differences (P < 0.05) between the 3- and > 12-h treatments, and between the 6- and > 15-h treatments. This suggests that the retention time does not exceed 3 h for 20-day-old dung and that it exceeds 9 but not 15 h for 36- to 40-day-old dung, although there is some uncertainty for the 20-day-old dung because there were only a few observations.

On the basis of all the data for the 20-day-old dung, the gut load of dung for *L. festivus* was  $0.06 \text{ g g}^{-1}$  ashfree dry weight of worm, which was significantly different from the above-mentioned value, and for 36- to 40-day-old dung, calculated from the values obtained after more than 12 h of exposure,  $0.11 \text{ g g}^{-1}$  ash-free dry weight, which was not significantly different from the above-mentioned value. A similar analysis for *L. castaneus* showed that the retention time exceeded 3 h but barely reached 6 h, irrespective of the age of the dung. The gut loads based on the results after 3 h of exposure were  $0.08 \text{ g g}^{-1}$  ash-free dry weight for 20-day-old dung and  $0.09 \text{ g g}^{-1}$  ash-free dry weight for 36-day-old dung.



**Fig. 1.** The amounts of chromic oxide marked dung in *Lumbricus festivus* after different durations of exposure to 20-day-old dung ( $\bullet$ ;  $\star$ , mean for each duration of exposure) or 36- to 40-day-old dung ( $\triangle$ , experiment 1;  $\Box$ , experiment 2;  $\star$ , mean of the two experiments for each duration of exposure); *AFDW*, ash-free dry weight





Fig. 2. The amounts of chromic oxide marked dung in *Lumbricus* castaneus after different durations of exposure to 20-day-old dung ( $\odot$ );  $\star$ , mean for each duration of exposure for both dung ages



Fig. 3. Frequency diagram showing the percentage of dung in defaecated material of *Lumbricus festivus* 

These two figures were not significantly different from the above-mentioned gut load of dung for *L. castaneus*.

Figure 3 shows that dung was mixed with variable amounts of other material during ingestion by *L. festivus. L. castaneus* was similarly variable (results not shown). Figure 4 indicates the admixture of soil with the dung ingested by *L. festivus.* 



Fig. 4. The relationship between dry weight and ash content of defaecated material of *Lumbricus festivus* collected below cowpats. Data for the dung and the soil are also shown

# Discussion

The results show that *L. festivus* and *L. castaneus* ingest dung but that dung constitutes only part of the material found in the gut. Furthermore, in *L. festivus* the gut loads and ash percentages in the faeces are higher for worms from free soil than for those found below cow pats. The comparison of gut loads on a weight basis is inadequate, because of differences in the density of the material, and comparisons should be made on a volume basis.

The retention time of food differs between the two species; a maximum of 15 h was observed for *L. festivus* eating 36- or 40-day-old dung, and 6 h at the for *L.* castaneus. The food-retention time for *L. festivus* was within the values determined for other *Lumbricus* spp., 8 h for *L. terrestris* at 25 °C (Hartenstein and Amico 1983) and 12-24 h for *L. rubellus* (Piearce 1972). The value for *L. castaneus* was closer to that found for *Eisenia fetida*, 2.5 h at 25 °C (Hartenstein et al. 1981).

L. festivus seems to exploit dung of different ages in different ways, since the gut load, and probably also the retention time, differed in relation to dung age. This is presumably related to the preferential aggregation of L. festivus below dung older than 20 days (Holter 1979, 1983). Differences in gut load have previously been reported for E. foetida, L. rubellus, and L. terrestris eating different food (Hartenstein et al. 1981; Shapitalo et al. 1988).

Piearce (1978) found variable amounts of inorganic material in the faecal output of six different species from a pasture. Kaplan et al. (1980) showed that the presence of soil, or at least some inorganic soil component, stimulated the early onset of sexual maturity in *E. fetida*. Likewise, I found different amounts of soil in the faecal output from *L. festivus* and *L. castaneus* when consuming

cow dung. These findings suggest that mineral particles are a necessary component of earthworm food.

From the mean gut contents of dung, the gut load of total ingested material and the retention time, it is possible to calculate both the amount of dung consumed and the total consumption per day for worms beneath pats. The dung consumption for L. festivus eating 36-day-old dung was calculated as 0.16 g dry dung g ash-free dry weight of worm  $day^{-1}$  at the maximum retention time of 15 h. The ash-free dry weight contributed 12.2% of the live weight of L. festivus, hence 1 g live L. festivus consumed 0.02 g dry dung day<sup>-1</sup>. This represented 0.08 g dung day<sup>-1</sup> (25% dry matter in dung). From this, the total consumption per day was calculated as 0.03 g dry material  $g^{-1}$  live weight for L. *festivus*. Similar calculation showed that L. castaneus consumed 0.15 g dung day<sup>-</sup> and 0.07 g dry material  $day^{-1}$ , when the retention time was the maximal 6 h.

Comparable consumption rates calculated on the basis of gut load and retention time were 0.04 g g<sup>-1</sup> live weight for *L. rubellus* in a woodland (Piearce 1972), 0.07 g g<sup>-1</sup> live weight for *L. terrestris* in reclaimed peat (Curry and Bolger 1984), and 0.45 and 0.77 g g<sup>-1</sup> live weight for *L. terrestris* in grassland soil and silt loam, respectively (Hartenstein and Amico 1983).

The detritivore earthworm fauna of the field station (Bøgemosen) in the autumn of 1987 totalled approximately 75 g m<sup>2</sup>. The species were L. festivus, L. rubellus, L. castaneus, and a few L. terrestris, with the two former dominant. Using the above consumption rates, the consumption of dung by the field populations was calculated as 10-15 t ha<sup>-1</sup> year<sup>-1</sup>, assuming that the earthworms were active for 180 days per year (approximately the period that cattle graze the grass in Denmark). This corresponds to the amount of dung produced by two or three dairy cows, or a little above the normal stocking rate. It is concluded that earthworms in Bøgemosen are able to consume all the dung produced by the livestock. However, these calculations have their limitations and the actual values must depent on the species present, the age of the dung, and also on abiotic factors such as soil temperature and water content.

Holter and Hendriksen (1988) measured the respiratory loss and bulk transport of organic matter from dung pats. They reported that both processes were important, and that the transport process contributed 54-75% to the disappearance of the dung pats. They proposed that earthworms were the main form of transport.

## References

- Barley KP (1959) The influence of earthworms on soil fertility. II. Consumption of soil and organic matter by the earthworm *Allolobophora caliginosa* (Savigny). Aust J Agric Res 10:179-185
- Curry JP, Bolger T (1984) Growth, reproduction and litter and soil consumption by *Lumbricus terrestris* L. in reclaimed peat. Soil Biol Biochem 16:253-257
- Dunn OJ (1964) Multiple contrasts using rank sums. Technometrics 6:241-252
- Guild WJMcI (1955) Earthworms and soil structure. In: Kevan DKMcE (ed) Soil zoology. Butterworth, London, pp 83-98
- Hartenstein R, Amico L (1983) Production and carrying capacity for the earthworm *Lumbricus terrestris* in culture. Soil Biol Biochem 15:51-54
- Hartenstein F, Hartenstein E, Hartenstein R (1981) Gut load and transit time in the earthworm *Eisenia foetida*. Pedobiologia 22:5-20
- Hendriksen NB (1991) Consumption and utilization of dung by detritivorous and geophagous earthworms in a Danish pasture. Pedobiologia 35:65-70
- Holter P (1973) A chromic oxide method for measuring consumption in dung-eating *Aphodius* larvae. Oikos 24:117-122
- Holter P (1979) Effect of dung-beetles (*Aphodius* spp.) and earthworms on the disappearance of cattle dung. Oikos 32:393-402
- Holter P (1983) Effect of earthworms on the disappearance rate of cattle droppings. In: Satchell JE (ed) Earthworm ecology. Chapman and Hall, London, pp 49-57
- Holter P, Hendriksen NB (1987). Field method for measuring respiratory loss and bulk export of organic matter from cattle dung pats. Soil Boil Biochem 19:649-650
- Holter P, Hendriksen NB (1988) Respiratory loss and bulk export of organic matter from cattle dung pats: A field study. Hol. Ecol 11:81-86
- Kaplan OL, Hartenstein R, Neuhauser EF, Malleski M (1980) Physicochemical requirements in the environment of the earthworm *Eisenia foetida*. Soil Biol Biochem 12:347–352
- McGinnis AJ, Kasting R (1964a) Chromic oxide indicator method for measuring food utilization in a plant-feeding insect. Science 144:1464-1465
- McGinnis AS, Kasting R (1964b) Colorometric analysis of chromic oxide used to study food utilization by phytophagous insects. Agric Food Chem 12:259-162
- Piearce TG (1972) The calcium relations of selected Lumbricidae. J Anim Ecol 41:167-188
- Piearce TG (1978) Gut contents of some lumbricid earthworms. Pedobiologia 18:153-157
- Satchell JE (1967) Lumbricidae. In: Burges A, Raw F (eds) Soil biology Academic Press, London, pp 259-322
- Shapitalo MJ, Protz R, Tomlin AP (1988) Effect of diet on the feeding and casting activity of *Lumbricus terrestris* and *L. rubellus* in laboratory culture. Soil Biol Biochem 20:233-237
- Van Soest PJ (1982) Nutritional ecology of the ruminant. O and B Books, Corvallis, Oregon
- Zar JH (1984) Biostatistical analysis. Prentice-Hall, Englewood Cliffs, New Jersey