Ann. appl. Biol. (1990), **116**, 169-176
Printed in Great Britain

Studies on *Artioposthia triangulata* (Dendy) (Tricladida: Terricola), a predator of earthworms

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(Accepted 27 October 1989)

Summary

Observations had linked the disappearance of earthworms from a grass field to the presence of *Artioposthia triangulata*. An experiment demonstrated that this land planarian could severely and quickly reduce numbers of four earthworm species. Two different approaches to sampling *A. triangulata* were investigated on a grassland site. The first used four different trap types (wood, ceramic tile, 'corriboard' plastic and 5 mm polystyrene beneath a ceramic tile) measuring 15 cm × 15 cm which were compared over a period of 18 wk. The second used one, or two, applications of dilute formalin solution to quadrats followed by counting the residual planarians by hand-sorting the soil beneath the quadrat to a depth of 30 cm. The polystyrene traps were the most effective for detecting the presence of *A. triangulata*. No planarians were ever found by hand-sorting after two formalin applications and it was concluded that formalin sampling provided a good estimate of the population density. Counts under traps were not related to absolute estimates of population densities so polystyrene type traps should only be used for detection of the planarians. A multiple regression relationship of *A. triangulata* weight over time in 5% formalin is presented and allows the original liveweight to be calculated from the weight in preservative. Planarian and earthworm population densities in eight fields were sampled and the planarians were found to be randomly distributed. Counts of planarians in 0.25 m^2 quadrats were not related to numbers of earthworms but overall field population densities were. It is concluded that *A. triangulata* is a severe threat to the earthworm populations of Northern Ireland.

Key words: Artioposthia triangulata, earthworms, predation, sampling, distribution, terrestrial planarian, Northern Ireland

Introduction

Artioposthia triangulata (Dendy) is a land planarian of the family Geoplanidae Stimpson originally recorded from the Christchurch area of New Zealand (Dendy, 1984) and can grow to 15 cm and weigh in excess of 2 g (R. P. Blackshaw, unpublished data). It was first discovered in Northern Ireland in the early 1960s (Willis & Edwards, 1977) and has subsequently been reported from many areas (Anderson, 1986 and unpublished records).

Willis & Edwards (1977) were able to demonstrate that mature *A. triangulata* fed on

Table 1. *Earthworm numbers recovered from 20 × 0.5 m² quadrats in grassland at Newforge Lane, Belfast with* A. triangulata *present at the site*

Sampling date	Population
March 1984	550
March 1985	735
March 1986	345
October 1986	530
March 1987	125
December 1988	0

earthworms in Petri dishes in the laboratory. They noted that the planarians wrapped themselves around the earthworms and killed them through the secretion of digestive juices. The resultant liquid was taken in to the planarian mouth. However, it was not until observations linked a marked decline in numbers of earthworms with activity of planarians in a field (Table 1) that they were regarded as being more than a curiosity. If they are able to deplete earthworm populations rapidly, their widespread distribution might indicate a developing ecological problem. An experiment testing *A. triangulata* predation of earthworms in a soil habitat is reported in this paper.

The planarians are most commonly seen under stones, dead wood or other surface debris but have also been found under traps set out for slugs (Anderson, 1986). Blackshaw (1989) recovered them whilst using a formalin expellant for earthworm sampling. These observations suggested two different approaches to the problem of sampling for *A. triangulata* and this paper reports the results of field investigations into the development of appropriate sampling techniques and their subsequent use in preliminary distribution studies.

Materials and Methods

Predation of earthworms

Tubes of 'Corriboard' (Killyleagh Box Company) 15 cm × 15 cm × 80 cm were filled to 10 cm from the top with a mixture of 10% heat sterilised horse manure in heat sterilised loam soil. Sufficient water was added to make the mixture moist throughout and the tubes were placed on wet capillary matting which was kept watered during the experiment. Earthworms were than added so that each tube contained a known number of one species viz. *Apporectodea caliginosa* (5), *Eisenia fetida* (5), *Lumbricus rubellus* (3) or *L. terrestris* (2). Corriboard lids were positioned on the soil surface to provide a shelter and help retain moisture. Two tubes of each species were randomly allocated to each of five blocks.

After 7 days, two *A. triangulata* were added to one of the tubes containing each species in each block by placing them on the soil surface below the lids. The tubes were left undisturbed for a further 3 wk before being opened. The number of worms in each tube was recorded.

Comparison of sampling methods

An area of established grass at Newforge Lane, Belfast (Grid ref. 329699), that had previously been used for an investigation into the effects of a calcareous seaweed product on earthworms (Blackshaw, 1989) was selected as a study site because it was known to harbour *A. triangulata*.

The site had been arranged in six blocks of five treatments (control, lime applied at 317 or 633 kg ha^{-1} yr^{-1} or calcareous seaweed applied at 317 or 633 kg ha^{-1} yr^{-1}) and had received applications each October from 1983 to 1986. The plots measured 5 m × 6 m.

On 15 April 1987, one trap consisting of a 15 cm × 15 cm flat sheet of each of four types (plywood, ceramic tile, corriboard plastic and 5 mm polystyrene sheet on the underside of a ceramic tile) was randomly placed in the corners of the 20 plots in four of the blocks. The grass was trimmed before placement and the traps were secured with tent pegs to avoid disturbance by birds. The traps were examined every 7 days for 18 wk. Any planarians found were counted, removed and released at the centre of the plot. Counts were transformed ($\log_e (n+1)$) and analysed with respect to trap type, sampling date and previous plot treatment.

On 9 June 1987, a single quadrat (0.5 m × 0.5 m) was randomly positioned in each of the 10 plots in the two blocks that had not yet been sampled and 4.5 litres of 0.5% formalin solution applied. After 20 min, the planarians on the soil surface were counted. The soil beneath the quadrats was then dug out to a depth of 30 cm, taken to the laboratory and hand-sorted to estimate the numbers of planarians not recovered by the formalin method.

On the same day, a further 10 quadrats (0.5 m × 0.5 m) were marked in the same 10 plots by sinking a steel frame 5 cm into the soil as an impediment to lateral surface movement by the planarians. The grass inside each quadrat was trimmed and a sheet of polystyrene, protected by hardboard, was positioned so as to completely cover the enclosed area. Externally, at each corner of the quadrat, one 15 cm × 15 cm trap of each of the four types described above was positioned on a random basis. After 7 days, the numbers of planarians on the soil surface under the polystyrene sheets within the quadrats and under the four smaller traps were recorded. The traps were then removed and formalin solution (0.5%) applied to the quadrats in two applications (4.5 litres each) 20 min apart; planarian counts were recorded separately after each application. In one block, the soil beneath the quadrats was removed to a depth of 30 cm for hand-sorting in the laboratory. At the end of this process, all the quadrats were moved and re-established. The sequence was repeated for four consecutive weeks and the counts for each of the four 15 cm × 15 cm trap types were compared with the total number obtained from the adjacent quadrat.

Weight change in formalin

Because the use of formalin may cause changes in bodyweight and make it difficult to estimate biomass, changes in planarian bodyweight with time in preservative were observed. A 5% formalin solution was made up as a preservative and 22 *A. triangulata* were washed, dried on paper towels, weighed and then placed in solution in individual containers. After 24 h, the planarians were taken from the preservative, dried and weighed before being replaced. This process was repeated after 2, 16, 30 and 64 days. A linear regression analysis of weight in preservative on original liveweight was carried out for each observation time.

Spatial distribution

Earthworm and planarian populations were sampled from 8 grass fields with two applications of formalin to 20 randomly placed quadrats (0.5 m × 0.5 m) in each field as described above. The earthworms and *A. triangulata* which emerged were collected and counted. For each site, the mean and variance of the planarian counts were calculated. The relationship between the numbers of planarians and earthworms was examined using the quadrat counts.

Results

Predation on earthworms

The data were analysed by considering the number of worms surviving as a proportion of the original number placed in each tube (Table 2). The planarians reduced ($P < 0.01$) the

Table 2. *Mean proportions of initial populations of earthworms recovered from experimental soil columns in the presence or absence of planarians. Standard deviations in parentheses*

	A. triangulata without	*A. triangulata* with
A. caliginosa [a]	0.80 (0.25)	0.32 (0.27)
E. fetida [a]	0.50 (0.28)	0.04 (0.09)
L. rubellus [b]	1.00 (0.00)	0 (0.00)
L. terrestris [c]	1.00 (0.00)	0.10 (0.23)

a, b, c — from an original 5, 3 and 2 worms respectively.

Table 3. *Mean of $\log_e (c + 1)$ transformed counts of planarians (untransformed data in parentheses) taken with 15 cm × 15 cm traps and the proportion of samples with positive counts (n = 360)*

	Wood	Ceramic tile	Corriboard plastic	Polystyrene/ ceramic	Standard error
Average catch	0.11[c] (0.16)	0.18[b] (0.28)	0.19[b] (0.29)	0.40[a] (0.71)	0.027 (0.051)
Proportion of positive counts	0.14	0.22	0.24	0.45	—

Means followed by a different letter are significantly different ($P < 0.001$).

numbers of all four species of earthworm over the three week period. There was no evidence of differential mortality but this may have been due to the small numbers of earthworms and planarians used in the experiment.

Comparison of sampling methods

There were no significant effects on numbers of *A. triangulata* taken from the 15 cm × 15 cm traps which could be attributed to either the treatments or the block design of the preceding experiment but there were significant differences ($P < 0.01$) between numbers trapped on different dates.

The different types of trap showed different degrees of effectiveness. The polystyrene covered tiles trapped significantly more ($P < 0.001$) planarians than any of the others and fewer were found under the wooden straps than either the ceramic tiles or the corriboard plastic (Table 3). This was reflected in the proportion of samples from the different types of trap that contained planarians.

The various combinations of formalin applications, hand-sorting and the use of a polystyrene sheet as a trap within quadrats gave rise to different counts (Table 4). The only time hand-sorting recovered any planarians was when it followed a single formalin application. For individual quadrats, the proportion of the total count trapped by the polystyrene sheet ranged from 0 to 1 and averaged 0.23 with a standard error of 0.343. Where two formalin applications were made the second only yielded a total of five individuals compared to 139 with the first.

The average population density within each quadrat was 19.4 m^{-2}. The 15 cm × 15 cm traps outside the quadrats gave average population density estimates as follows: wood, 7.8 m^{-2}; ceramic tile, 11.1 m^{-2}; corriboard plastic, 12.2 m^{-2}; polystyrene/ceramic, 36.7 m^{-2}. Quadrat counts were not significantly correlated with adjacent wooden ($r = -0.047$), ceramic tile ($r = -0.294$), corriboard plastic ($r = -0.076$) or polystyrene/ceramic ($r = -0.242$) traps.

Table 4. *Numbers of planarians recovered from 0.5 m × 0.5 m quadrats using different sampling methods applied sequentially*

Polystyrene sheets	Formalin 1st	Formalin 2nd	Hand Sorting	n
*	14	*	2	10
14	50	3	*	20
36	89	2	0	20

* sampling procedure not carried out

Table 5. *Relationships between the weight of* A. triangulata *in formalin and initial liveweight after various periods of immersion (n = 22)*

Time (days)	Regression coefficient	regression constant	Correlation coefficient
1	1.251	−0.004	0.99
2	1.198	−0.006	0.99
16	1.169	−0.006	0.99
30	1.121	−0.010	0.99
64	1.110	0.003	0.99

Weight change in formalin

The weight of *A. triangulata* increased by approximately 25% over the first 24 h in 5% formalin; weight gradually declined thereafter but never returned to the original liveweight (Table 5).

A multiple regression model was derived for estimating initial liveweight from the weight of a preserved planarian and the time spent in 5% formalin. The best fit showed that the relationship preserved weight = (113.04 × liveweight) − (7.36 × days in preservative) + 2.60 accounted for 99.2% of the variation in the data.

Spatial distribution

Logarithmic transformations of the mean and variance associated with each of the 8 populations of *A. triangulata* were taken and plotted against each other (Fig. 1) and the relationship between them calculated as: $\log_e$ (variance) = 1.071 × $\log_e$ (mean) + 0.034 r = 0.96. The gradient of this regression represents an index of aggregation (Taylor, 1961) and suggests that the distribution of the planarians was approximately random.

Numbers of planarians and earthworms from individual quadrats showed (Fig. 2) no correlation at this scale, but the mean field populations were significantly related (N_p = 0.117 N_e + 1.167, r = 0.89; where N_p and N_e refer to *A. triangulata* and earthworms per 0.25 m^2 respectively).

Discussion

Out of 75 earthworms in tubes containing planarians, only 10 survived for three weeks compared with 59 in the controls. This provides evidence that *A. triangulata* can kill earthworms in a soil environment and, potentially, could be responsible for a rapid decline

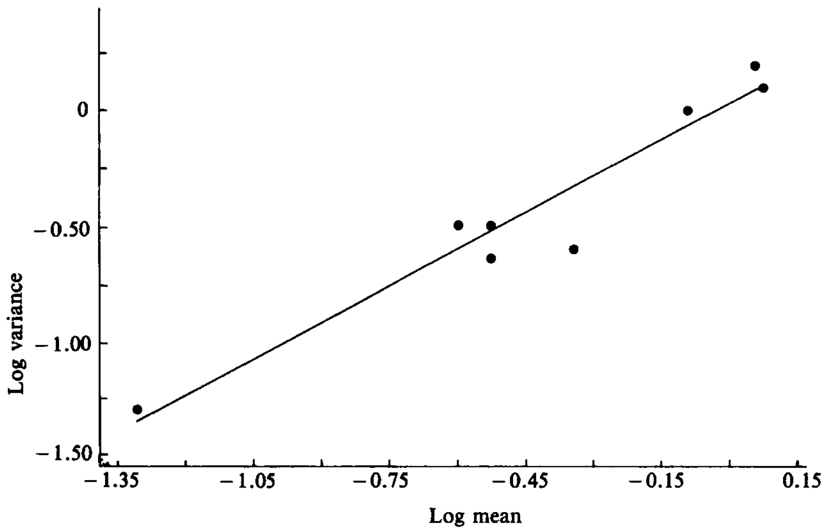

Fig. 1. Relationship between $\log_e$ variance and $\log_e$ mean for *Artioposthia triangulata* population densities from 8 fields.

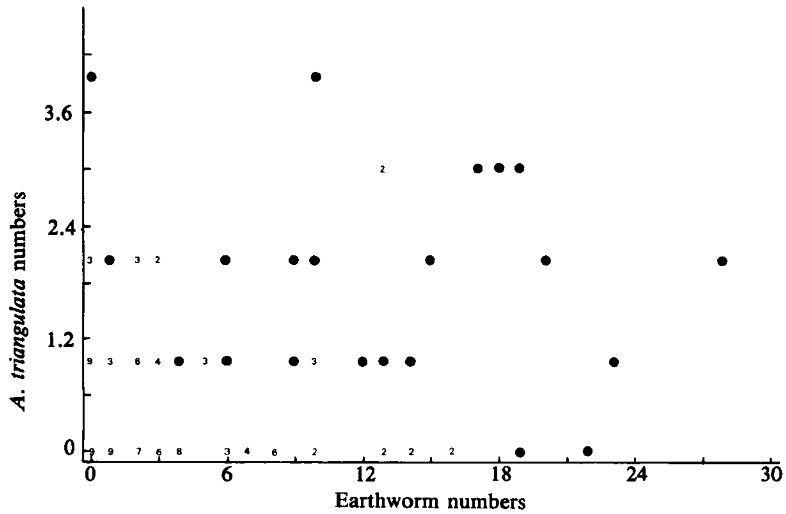

Fig. 2. Earthworm and *Artioposthia triangulata* counts in 0.25 m^2 quadrats. Numbers represent coincident points; 9 may equal 9 or more.

in their numbers. This supports the earlier observations (Table 1) where an earthworm population was almost eliminated from a grass field over a period of only a few years.

Greater catches of *A. triangulata* were obtained through the use of polystyrene/ceramic type traps than with any other type examined and a greater proportion of these traps yielded planarians each week (Table 3). Thus, this type of trap is more effective for detecting planarians than the others.

If it is assumed that those planarians found under the polystyrene sheets within the quadrats would have been expelled by formalin sampling anyway, a single formalin application would be sufficient to obtain a good estimate of the population (Table 4). A second application recovered all individuals present within the profile of a quadrat to a depth of 30 cm, the deepest recorded observation (Willis & Edwards, 1977).

The variable proportions of the total population of *A. triangulata* taken under the polystyrene sheets within quadrats and the poor correlations between 15 cm × 15 cm traps and adjacent quadrat counts demonstrated the unreliability of trapping as a means of estimating the population density.

Although not correlated, the mean number of *A. triangulata* m^{-2} taken under polystyrene/ceramic traps outside the quadrats during the formalin studies was almost double that revealed by trapping and two formalin applications within the quadrats. The other trap types examined yielded lower population densities than the quadrats. This suggests that the sampling ambit of the polystyrene/ceramic traps was greater than their dimensions and that they encouraged planarian congregation. This may have been due to the insulating properties of polystyrene.

Whatever the reason for their effectiveness, polystyrene/ceramic type traps provide a simple method for the detection of *A. triangulata*, but they are unreliable for estimates of population densities. Absolute estimates of numbers can be obtained through the use of two applications of dilute formalin solution. This approach has the additional benefit of also providing an estimate of the numbers of the planarian prey – earthworms (Raw, 1959).

The use of formalin to sample planarian populations may affect biomass assessments, even with concentrations as low as 0.5%. Placing samples in 5% formalin for later weighing and back-calculation of liveweight can be contemplated. If this procedure were adopted for formalin extracted individuals, it would be advisable to delay weighing and use the multiple regression correction equation rather than weighing as soon as possible, so that error can be minimised.

The planarians were found to occur randomly within the fields sampled in this study and their numbers were not related to the numbers of earthworms within individual quadrats. Along with the enhanced catch per unit area for the polystyrene/ceramic traps, these results suggest an organism that is relatively mobile.

There did appear, however, to be a relationship between mean population densities of planarians and earthworms within fields. This is probably not a constant relationship and will have been influenced by the selection of study sites known to have *A. triangulata* present. One, for example, lacked earthworms and had only a very sparse population of planarians, presumably because of a shortage of food. A more detailed study of this relationship may provide useful information on spread to and colonisation of new areas.

The picture that has emerged from these initial studies is of an organism that may be capable of inflicting severe losses on indigenous earthworm populations to the point of elimination. When this occurs it is possible that they are sufficiently mobile to disperse and locate new concentrations of earthworms. *A. triangulata* is now found right across Northern Ireland and into Donegal. If Willis & Edwards (1977) are correct, the planarian has achieved this spread within the last 25 to 30 years. There is no evidence, as yet, for any natural regulation of *A. triangulata* numbers other than shortage of food, so the outlook for Northern Ireland's earthworm fauna may be bleak.

Acknowledgements

I thank Messrs J. P. Moore, J. F. Calvert and C. C. Woods for their technical assistance.

References

Anderson, R. (1986). The land planarians of Ireland (Tricladida: Terricola) a summary of distribution records. *Irish Naturalists Journal* **22**, 141-146.

Blackshaw, R. P. (1989). The effects of a calcareous seaweed product on earthworms in a grassland soil. *Biological Agriculture & Horticulture* **6**, 27-33.

Dendy, A. (1894). Notes on New Zealand Land Planarians. Part 1. *Transactions of the New Zealand Institute* **27**, 177-189.

Taylor, L. R. (1961). Aggregation, variance and the mean. *Nature, London* **189**, 732-735.

Raw, F. (1959). Estimating earthworm populations by using formalin. *Nature, London* **184**, p. 1661.

Willis, R. J. & Edwards, A. R. (1977). The occurrence of the land planarian *Artioposthia triangulata* (Dendy) in Northern Ireland. *Irish Naturalists Journal* **19**, 112-116.

(Received 12 June 1989)