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The effect of earthworms and snails in a simple plant community

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Abstract. Snails and earthworms affected the dynamics of a simple, three-species plant community, in the Ecotron controlled environment facility. Earthworms enhanced the establishment, growth and cover of the legume *Trifolium dubium*, both via the soil and interactions with other plant species. Worms increased soil phosphates, increased root nodulation in *T. dubium*, and enabled *T. dubium* seedlings to establish in the presence of grass (*Poa annua*) litter, by increasing soil heterogeneity. Worms also buried the seeds of *Poa annua* and *Senecio vulgaris*, reducing the germination of new seedlings. Snails reduced nitrogen-fixing *Trifolium dubium* and increased cover of plant litter, thereby reducing ammonia-nitrogen concentrations in the soil. These effects and their interactions demonstrate that the detritivore food chain, and earthworms in particular, cannot be ignored if we are to understand the spatial and temporal dynamics of plant communities.

Key words: Ecotron – *Trifolium dubium* – Earthworms – Molluscs – Plant community dynamics.

Following Darwin's (1837, 1881) pioneering studies on earthworms a voluminous literature records the effects of earthworms on the physical and chemical structure of soil (see Lee 1985 for review; Satchell 1983). Earthworms increase the productivity of plant communities, in both controlled (Hopp and Slater 1948; van Rhee 1965) and field conditions (Stockdill 1966; Hoogerkamp et al. 1983, Sears and Evans 1953). Interrelations between the activities of earthworms, the soil and plant growth are, however, complex, and involve changes in the rates of turnover of major nutrients such as nitrogen and phosphorus (Barley and Jennings 1959; Aldag and Graff 1975; Lunt and Jacobson 1944; Sharpley and Syers 1976, 1977). Suggested mechanisms (Mackay et al. 1983) include enhanced microbial activity, commonly found in earthworm casts (Barley and Jennings 1959; Parle 1963; Jeanson 1960), increases in the rates of removal and incorporation of dead material from the soil surface (Darwin 1881; Stockdill 1966, 1982), and changes to the soil structure (Tisdall 1978).

In recent years the effects of earthworms on plant communities have been somewhat neglected, despite early examples of increases in the proportion of *Trifolium* in plant communities to which earthworms had been added (Hopp and Slater 1948; van Rhee 1965). The implications of such observations for the structure of plant communities remain poorly understood.

The role of herbivory by molluscs in structuring plant assemblages has received considerable attention (Dirzo 1985; Edwards and Gillman 1987; Cottam 1986; Rees and Brown 1992). By selectively grazing certain plant species, particularly seedlings (which they may kill), molluscs alter the species composition and relative abundances of plant communities (Edwards and Gillman 1987). But are molluscs more or less important in their effects than earthworms? What sort of interaction, if any, might be expected between these two very different groups of animals in determining the dynamics of plant communities?

The paper describes a series of experiments to investigate how earthworms and snails affect the composition of a simple plant community, both through their direct effects on the competitiveness of the different species and through their interactions with the rhizosphere.

Materials and methods

Ecotron experiment

The main experiment was conducted in the Ecotron at the NERC Centre for Population Biology at Silwood Park. This controlled environment facility simulates natural environments in diurnal light: dark cycle, temperature and rainfall. A full description of the Ecotron is given in Lawton et al. (in press).

The experiment used 8 of the 16 available chambers, with two replicates of each of four treatments. All the chambers contained the same number and distribution of individuals of each of three plant species; *Poa annua* (Graminae), *Senecio vulgaris* (Compositae) and *Trifolium dubium* (Leguminoseae). The four treatments consisted of adding either no animals, snails (*Helix aspersa*), casting species of worms (*Lumbricus terrestris* and *Aporrectodea* spp., mostly *A. longa*) or a combination of both snails and worms. Environmental conditions in the chambers are summarised in Table 1.

Communities were established in large (1.3 m × 0.85 m × 0.37 cm deep) containers with free drainage, with one container

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Table 1. Climate data for the Ecotron between July 1991 and April 1992

Temperature Smooth diurnal cycle	Maximum: 20° C (at 1500 h) Minimum: 12° C (at 0600 h)
Photoperiod	16 h from 0430 to 2030 h (with a gradual dawn and dusk each of 60 min)
Light intensity	300 μm quanta $m^{-2} s^{-1}$
Relative humidity	Maximum: 70% (at 0600 h) Minimum: 58% (at 1500 h)
Rainfall	2 mm day^{-1} delivered over 5 min at the end of the photoperiod

placed in the centre of each walk-in Ecotron chamber. The centre of each container was occupied by a vertical pipe to improve the circulation of controlled environment air throughout the plant canopy (air at controlled temperature and humidity enters the chamber through the floor). The pipe covered an area of 0.28 m^2; the surface area of the plant community within each chamber therefore occupied 0.8 m^2 (Fig. 1).

Each container was set up with 10 cm washed gravel at the base topped with 0.26 m of a soil mixture consisting of 50:50 'Surrey Cricket Loam' (sterilised and screened) and '3, 2, 1 Top Dressing', (supplied by STABRITE, Basingstoke). The containers were inoculated with a 1-cm layer of unsterilised, sieved (to 2 mm) field soil (of a similar type to the mixture), buried 14 cm below the soil surface to provide a natural microflora, but to prevent the germination of seeds. The containers were covered with muslin and allowed to stand for 1 month until experimental seeds were sown on 26 June 1991. The initial density and distribution of seeds of the three plant species was the same in all the containers. Second-generation performance of *Senecio* growing with *Poa annua* is significantly improved if the distribution of the *Poa* is patchy in space, and that of the *Senecio* is random (Bergelson 1990). We distributed the grass and dicotyledons accordingly.

The surface of each container was divided into a 4 × 6 grid of 20 × 20 cm squares, producing a total of 20 experimental squares, the central 'hole' taking up an area equivalent to four squares (Fig. 1).

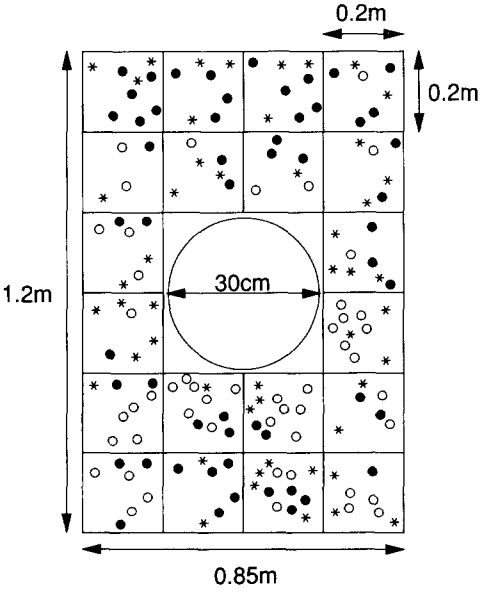

Fig. 1. Initial distribution of *Poa annua* (○), *Senecio vulgaris* (*) and *Trifolium dubium* (●) within the grid arranged on each Ecotron container. The *30-cm-diameter pipe* in the *centre* increases air flow over the community

The patchy distribution of *Poa* was determined using a negative binomial distribution with a mean of 3, and $k=1$, and the random distribution of the dicotyledons was determined by a Poisson distribution of the same mean. For each species the number of seeds in each grid square was randomly drawn according to the specified distribution. The spatial arrangement of the seeds within each grid square was randomly determined, and maintained over all the containers (Fig. 1). The overall density of plants was 203 m^{-2} (70 plants m^{-2} for *Senecio* and *Trifolium* and 63 plants m^{-2} for *Poa*).

The planting patterns were established by superimposing a 20 × 20 cm grid on the soil surface, within which 20 × 20 cm wooden squares were placed with 1-cm-diameter holes drilled at the previously determined positions of the plants. Between 5 and 10 seeds of each species were then dropped through the appropriate holes. Seedlings were thinned until a single adult plant remained in each position.

Treatments were randomly assigned to each chamber, and the animals introduced after 1 month. A total of 200 snails were collected from a garden in Bracknell, Berkshire, and after selection for a standard size, 25 were placed on the soil surface of appropriate containers. Worms were collected from Silwood Park using a 1% formalin solution and kept on moist tissue for 24 h to evacuate their gut contents. After selection for a standard size (7–10 cm long) 185 individuals in total (*Lumbricus terrestris* and *Aporrectodea* spp. in proportion to their abundance in the field collection), were introduced onto the soil surface of appropriate containers.

Percentage cover of each plant species (plus bare ground and litter) was estimated every 2 weeks by taking two photographs per container, centred directly above each end. Slides were then projected onto a 10 cm grid and the percentage of each square occupied by each species recorded by placing a 10 × 10 grid of 1 × 1 cm squares marked on acetate over each of the larger squares. Later estimates were made by eye, once it had been determined that this method gave consistent results. Data were arcsine transformed before analysis. Confidence limits shown on the figures were calculated from an analysis of variance on the untransformed data; we analysed the results using a repeated-measures analysis of variance.

The experiment ran for 9 months (from July 1991 to April 1992), at the end of which the following measurements were recorded.

Nodulation

Five *Trifolium* roots were taken from five separate plants randomly selected (by position) from each container. The roots were washed carefully, the number of nodules recorded and the roots dried and weighed, to produce a measure of the number of nodules per gram dry weight.

Soil analysis

Soil cores (5 cm wide × 15 cm deep) were taken from five random positions within each container, to estimate concentrations of available nitrogen, phosphorus and potassium. These analyses are simple, and for this reason, and because they were only made on soil at the end of the experiment, require caution in interpretation. Nitrogen was measured by extracting samples in 2M potassium chloride solution, and analysing for ammonia-nitrogen (Patton and Crouch 1977) and nitrates and nitrite (Nydhal 1976). Phosphate-phosphorus was measured using the 'molybdenum blue method' (Golterman et al. 1978), and a Technicon autoanalyser, (Technicon Instruments, Chertsey, Surrey). The potassium concentration was assessed using an atomic absorption spectrophotometer (Thermo Jarrell Ash. Corporation, Franklin, Massachusetts) after extraction in 1M ammonium nitrate.

Animals

Numbers of worm casts on the surface in each grid square were recorded at the end of the experiment. The containers were then

emptied and the contents sieved, to recover worms still present in the soil. All surviving snails were collected and counted.

Additional experiments

The effect of soil samples from each treatment on the germination of Trifolium dubium. Results showed a major effect of earthworms on the population dynamics of *Trifolium* (see below). A series of experiments was therefore carried out to determine the mechanism(s) involved. These involved testing the soil, the presence and absence of *Poa* litter (which built up in the ecotron experiments to form dense mats) and the role of worm casts on the germination and performance of *Trifolium*.

Soil cores (taken in the same manner as for the soil analysis at the end of the main experiment in April 1992) from each chamber were retained and used to fill circular petri dishes 9 cm in diameter × 1 cm deep (five dishes per container), and 50 seeds of *Trifolium* spread evenly on the surface. The dishes were stacked in five blocks, containing one replicate from each chamber; light was excluded by covering the stacks with aluminium foil, and the stacks stored at 21° C. The number of seeds that germinated in each dish was recorded every 2 days for 12 days, until all the seeds had germinated. The soil was kept moist by applying a fine mist of distilled water daily.

The effect of worm conditioned soil on the growth of Trifolium dubium. Worm casts and soil samples were collected from Ecotron containers with and without worms in April 1992 (at the end of the main experiment). Ten 9-cm flower pots were filled with each of the three 'soil' types; (i) worm casts, (ii) soil from containers with worms, (iii) soil from containers without worms. Ten seeds of *Trifolium dubium* were sown in the centre of each pot, and the seedlings thinned so that each pot supported only a single individual. The plants were left to grow in an unheated greenhouse for 6 weeks between May and July 1992, after which they were harvested, washed, and the dry weight recorded.

Seedbank. Ten randomly positioned soil cores (5 cm diameter × 15 cm deep) were taken from each container in April 1992 and divided into 24 small pots (4 cm × 4 cm × 6 cm); the pots were kept watered and stored in an unheated glasshouse for 1 month during July 1992. Every 3 days emergent seedlings were removed and their number and species recorded.

Worm-casts as safe microsites for the germination of Trifolium dubium. The same soil mixture as in the main ecotron experiment was used to fill 54 seed trays (20 × 40 × 5 cm). Two-thirds (36) of the trays were sown with *Poa* evenly distributed at a density of 70 seeds m^{-2}. The plants were left to grow for 8 weeks in April–June 1992, after which time *Poa* had flowered and begun to senesce. The *Poa* was then cut in all trays, and the litter was removed from 18 of the trays.

These manipulations generated 18 replicates of each of the following treatments:

1. Undepleted soil
2. Depleted soil − *Poa* litter
3. Depleted soil + *Poa* litter

Worm casts, collected from the ecotron experiment were mixed with distilled water and used to produce standard casts 5 cm in diameter by 2 cm deep. The same size moulds were used to produce 'false casts' of exactly the same dimensions from fresh soil mixture.

Six replicates of treatments 1–3 above were then allocated to receive either (a) worm casts, (b) 'false' worm casts or (c) no casts. For (a) and (b), eight casts (real or false) were placed evenly on the soil surface. Where litter was present half of each cast was buried beneath the litter.

All the trays were then watered and 100 *Trifolium* seeds were broadcast in 20 g of fine sand as evenly as possible over the surface. The trays were left for 8 weeks in an unheated glasshouse during July an August 1992. After 8 weeks all the vegetation was removed from the trays and the established *Trifolium* dried and weighed.

Results

Ecotron

Results are summarised in Figs. 2–5 and Table 2.

The three plant species flowered in the same order as they would have over the course of a natural spring and summer. *Senecio* was initially dominant (Fig. 2a), but died after flowering. *Poa* then became the dominant species (Fig. 2b), and as the live *Poa* died (Fig. 2c), it was replaced by *Trifolium* (Fig. 2d). The start of a second generation of *Senecio* appeared during the last weeks of the experiment (Fig. 2a). *Senecio* was the only species that produced distinct generations. Seedlings of *Poa* and *Trifolium* entered populations continuously from c. 60 days. Flowering individuals of both *Poa* and *Trifolium* were present from day 48 to the end of the experiment, with no significant differences detected in the number of flowers produced between any of the treatments. The presence of animals did not significantly affect either the abundance or generation time of *Senecio* (Fig. 2a).

Poa reached a peak of abundance in all chambers at c. 85 days. The plants then began to die (Fig. 2b), with a rise in *Poa* litter after this time (Fig. 2c). By 111 days the treatments began to diverge significantly in the abundance of *Poa* (Wilks lambda 0.006; Table 2). Containers with worms and no snails supported significantly less *Poa* than those containing snails ($P = 0.02$; Table 2). *Poa* continued to senesce until the end of the experiment (Fig. 2c) in all treatments, the litter beginning to disappear during the last month when *Senecio* began to reappear. Significantly less *Poa* litter was found in the two treatments without snails after day 192 compared to those containing snails ($P < 0.001$; Table 2).

Trifolium remained below c. 20% cover in all treatments for about the first 4 months (111 days) after which

Table 2. Probability values from a repeated-measures analysis of variance between days 111 and 161 (time period 1) and days 179 and 245 (time period 2)

	Probability of significant differences occurring among treatments.	Probability of significantly different temporal patterns among treatments. (Wilks lambda)
Time period 1		
Trifolium	0.044	0.000
Poa	0.156	0.006
Dead *Poa*	0.019	0.001
Trifolium flowers	0.246	0.520
Poa flowers	0.647	0.152
Time Period 2		
Trifolium	0.016	0.762
Poa	0.020	0.531
Dead *Poa*	0.000	0.313
Trifolium flowers	0.085	0.031
Poa flowers	0.086	0.347

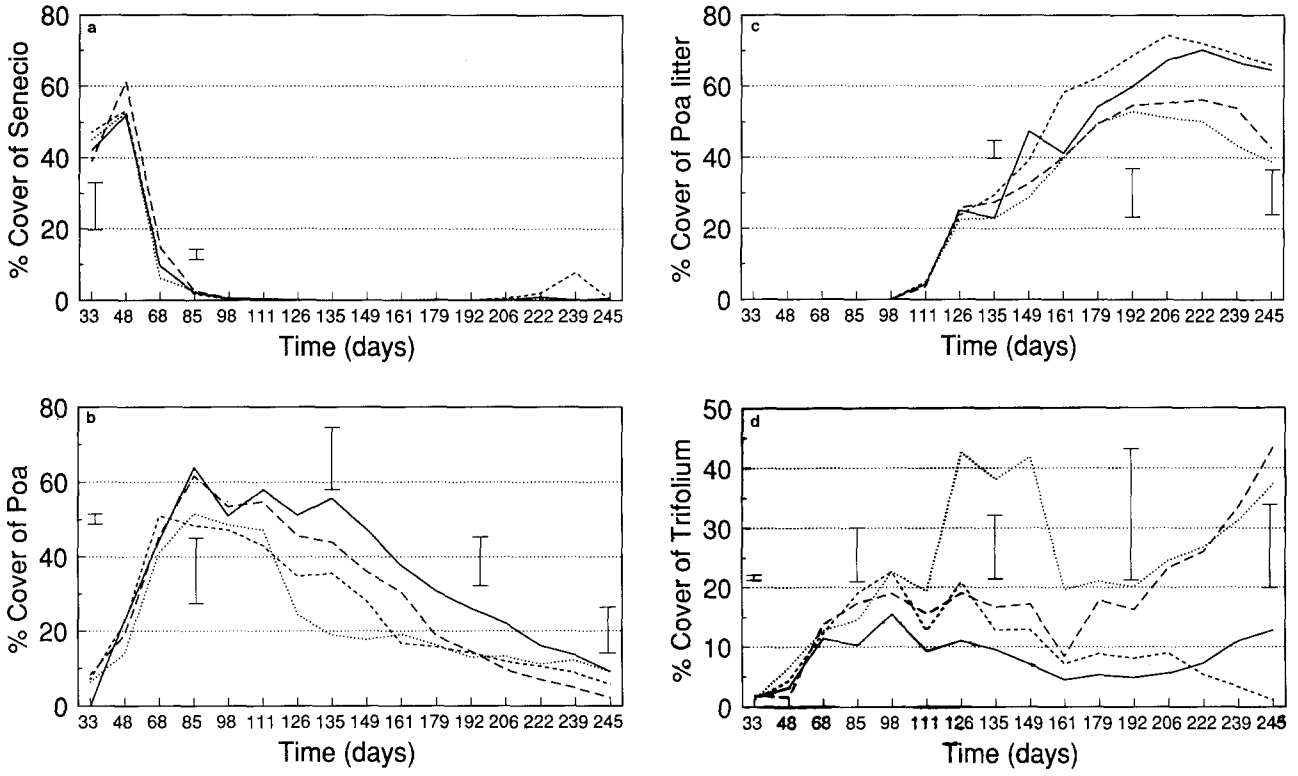

Fig. 2. **a** The mean percentage of the total area of the Ecotron container occupied by *Senecio vulgaris* in containers with no additional animals (——), worms (.....), snails (– – –) and both worms and snails (——). Illustrative 95% confidence intervals are shown. **b** The mean percentage of the total area of the Ecotron container occupied by *Poa annua* with no additional animals (——), worms (.....), snails (– – –) and both worms and snails (——). Illustrative 95% confidence intervals are shown. **c** The mean percentage of the total area of the Ecotron container occupied by dead *Poa annua* (litter) with no additional animals (——), worms (.....), snails (– – –) and both worms and snails (——). Ilustrative 95% confidence intervals are shown. **d** The mean percentage of the total area of the Ecotron container occupied by *Trifolium dubium* with no additional animals (——), worms (.....), snails (– – –) and both worms and snails (——). Illustrative 95% confidence intervals are shown

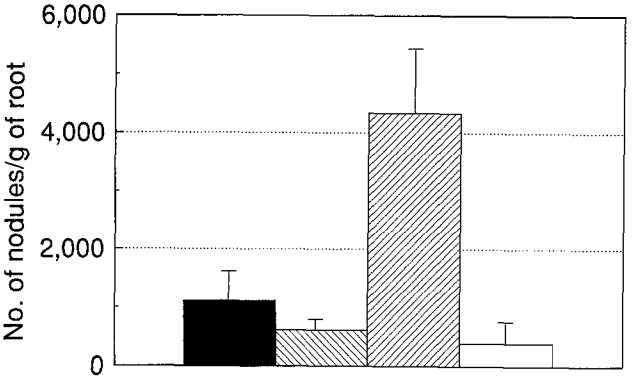

Fig. 3. Mean number of nodules (±95% CI) per g of *Trifolium dubium* root sampled from containers with no animals (□), snails (▨), worms (▨) and snails and worms (■)

the abundance of *Trifolium* in the worm/no snails treatment increased rapidly. Two weeks later the ecosystems containing worms/no snails supported double the cover of *Trifolium* compared to the other treatments. (Fig. 2d, Table 2.). This high cover was maintained for c. 2 months, after which the amount of *Trifolium* in all the chambers fell

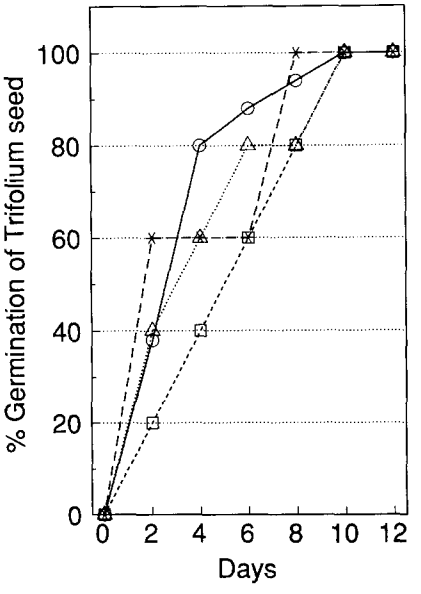

Fig. 4. Cumulative percentage germination of *Trifolium dubium* seeds on soil sampled from treatments with no animals (- x -), snails (-□-), worms (···△···) and snails and worms (—○—)

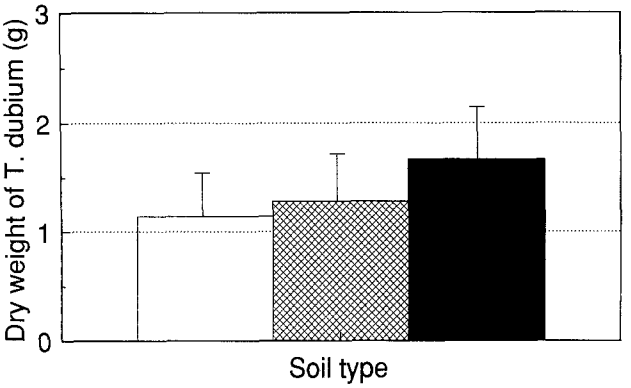

Fig. 5. Dry weight of *Trifolium dubium* (±95% CI) grown for 6 weeks on worm casts (■), soil sampled from treatments containing worms (▩) and soil sampled from treatments with no worms (□)

to approximately half that recorded during the previous 2 months. High abundance of *Trifolium* in the worm/no snails treatment coincides with the period when the cover of *Poa* was significantly reduced. *Trifolium* began to increase again around day 200 in the chambers without snails. The cover of *Trifolium* remained below 10% in the treatments containing snails until the last month of the experiment. It then began to rise in the chambers containing worms in addition to the snails, but disappeared from the snail-only treatments.

Nodulation

The number of nodules per gram dry weight of root in treatments containing worms as the only animals was 100 × more than the control treatments ($F=81.004$, $P<0.001$; 2-way ANOVA; Fig. 3). Root nodulation also increased in *Trifolium* grown in treatments containing snails without worms ($F=36.815$, $P<0.001$), and a significant interaction between worms and snails was detected ($F=48.438$, $P<0.001$).

Soil analysis

Data were log transformed and subjected to a two-way analysis of variance. There was a significant reduction in the availability of ammonia-nitrogen in treatments with snails, but not in treatments with worms. There was no significant interaction between the snails and worms on the availability of ammonia-nitrogen (2-way ANOVA; snail effect, $F=8.226$, $P=0.007$; worm effect, $F=0.778$, $P=0.384$; snail × worms interaction, $F=0.118$, $P=0.734$) (Table 3).

Available phosphates (Table 3) were significantly increased in the presence of worms, but unaffected by snails; no interaction between the two groups of animals was detected (2-way ANOVA: snail effect, $F=2.305$, $P=0.138$; worm effect, $F=9.739$, $P=0.004$; snail × worms interaction $F=0.064$, $P=0.802$). Animals (worms and/or snails) did not significantly affect the concentration of either nitrates (overall ANOVA: $F=0.959$, $P=0.432$, and all effects non-significant) or potassium (overall ANOVA $F=1.225$, $P=0.135$ and all effects non-significant) in the soil.

Table 3. Mean concentrations (mg/kg) (±95% CI) of ammonia–nitrogen and available phosphate in soil sampled from containers with no animals, snails, worms, and snails and worms (95% confidence intervals), at the end of the experiment

	Ammonia–nitrogen	Available phosphate
No animals	0.899 (0.439)	3.553 (1.480)
Snails	0.505 (0.352)	4.230 (0.584)
Worms	0.490 (1.101)	4.946 (1.249)
Snails and worms	0.601 (0.348)	5.523 (1.020)

Table 4. Mean numbers and (a) weight of worms and (b) diameter of snails recovered from containers at the end of the experiment (95% confidence intervals)

	Mean number of worms	Mean live weight of worms (g)
(a)		
Worms alone	57.50 (±49.49)	1.029 (±0.429)
Worms + snails	59.00 (±17.21)	0.734 (±0.973)
(b)		
	Mean number of snails	Mean diameter of snails (mm)
Snails alone	115.00 (±55.95)	16.52 (±0.839)
Snails + worms	101.50 (±89.95)	19.13 (±0.021)

Animals

There was no significant differences at the end of the experiment in the number of worm casts m^{-2} [worms only, $\bar{x}=818.12\pm132.05$ (±95% CI) and worms with snails $\bar{x}=810.00\pm130.75$] (1-way ANOVA; $F=0.025$, $P=0.223$). In all cases, the numbers of worms had fallen to approximately 25% of initial numbers. There were no significant differences in either worm numbers (1-way ANOVA; $F=0.015$, $P=0.913$) or worm weights (1-way ANOVA; $F=1.424$, $P=0.355$) between treatments (Table 4a). The proportions of *Lumbricus* and *Aporrectodea* did not appear to alter during the course of the experiment, although we did not measure this precisely.

There were large increases in numbers of snails (from 25 to c. 100 individuals per container) but numbers of snails were not affected by the presence or absence of worms (1-way ANOVA: $F=0.058$, $P=0.832$) Table 4b). Individual snails were significantly larger from treatments containing worms (1-way ANOVA: $F=179.03$, $P=0.006$).

Additional experiments

Soil type did not affect the germination of *Trifolium* (1-way ANOVA: $P=0.567$) (Fig. 4). Soil did, however, have a significant effect on the growth of this species (1-way ANOVA: $F=6.093$, $P=0.006$). *Trifolium* harvested from

pots containing only worm casts were significantly more productive (Tukey multiple-comparison test, $P=0.006$) than those from pots containing depleted soil. The least productive plants (Tukey multiple-comparison test, $P=0.030$) were those from pots containing depleted soil that had not supported any worms (Fig. 5).

Very few *Trifolium* seeds emerged from any of the soil cores tested, and this was not significantly affected by treatment. Most *Trifolium* seeds must have germinated rapidly in the Ecotron, as they did in our separate experiment (Fig. 4). However soil cores from treatments containing worms had significantly higher numbers of both *Senecio* (Fig. 6a) (1-way ANOVA: $F=3.524$, $P=0.024$) and *Poa* (Fig. 6b) (1-way ANOVA: $F=763$, $P<0.001$) seeds than did those from containers containing no animals or snails alone.

In the comparison of real and false worm casts, the presence of *Poa* either above (litter) or below ground (roots) significantly reduced the abundance of *Trifolium* (2-way ANOVA: $F=4.769$, $P=0.018$). There were no significant effects of false casts or real worm casts on the establishment of *Trifolium* in any of the treatments ($F=0.299$, $P=0.744$), and no interaction between the two factors ($F=1.036$, $P=0.409$). However, if the treatments containing *Poa* (both below ground (−litter), and above and below ground (+litter)) were compared, a marginally significant interaction between 'casts' (the presence, and type of cast) and litter was detected ($F=3.067$, $P=0.076$). In treatments containing above ground *Poa* (litter) the biomass of *Trifolium* was increased in the presence of either real worm casts ($P=0.041$, Tukey's miltiple-comparisons test) or simulated casts ($P=0.03$, Tukey's multiple-comparisons test), with no significant difference between the two (either real or false casts) ($P=0.918$, Tukey's multiple-comparisons test) (Fig. 7).

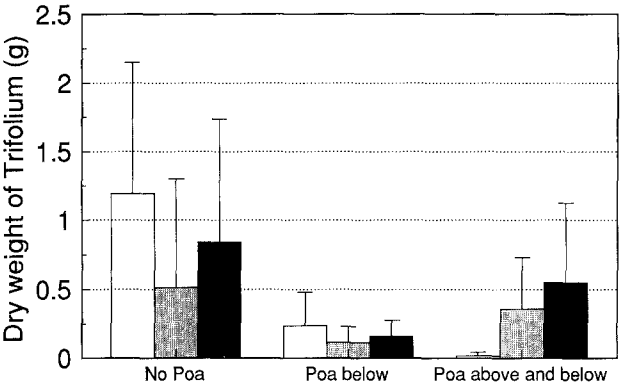

Fig. 7. Dry weight of *Trifolium dubium* (±95% CI) harvested from treatments with no *Poa annua*, and *Poa annua* below ground and both above and below ground, with no additional 'cast' treatment (□), with false casts (▨) and real worm casts (■)

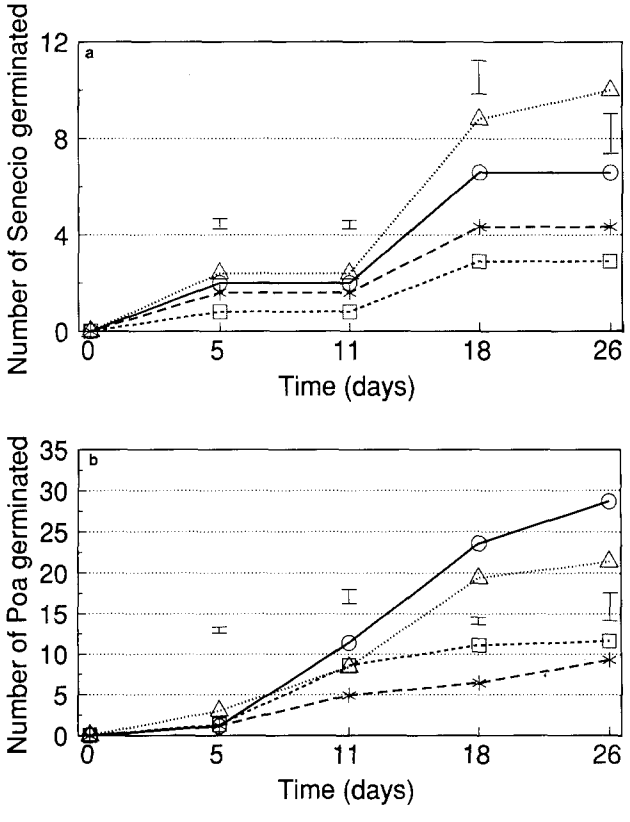

Fig. 6. Cumulative numbers of seeds germinating for **a** *Senecio vulgaris* and **b** *Poa annua* in soil cores sampled from Ecotron treatments containing no animals (- x -), snails (-□-), worms (···△···) and snails and worms (—○—). 95% confidence intervals are shown

Discussion

These are the first substantial results to emerge from the Ecotron (Lawton et al. in press), and accordingly they require cautious interpretation. There is still a great deal that we do not know about the processes going on in these simple communities. The soil, for example, is rather artificial, and poorly studied. Replication of treatments was also low.

Despite these uncertainties, there are strong, highly statistically significant effects of earthworms and snails on the dynamics of this simple plant community. In particular, containers with worms and no snails supported almost 3 times the amount of *Trifolium dubium* than did the other communities, in the first generation (between days 111 and 161). Although unexpected, these results are not unique. Similar increases in the amount of the related *Trifolium repens* in simple communities in the presence of earthworms were reported by Hopp and Slater (1948).

Earthworms influenced the performance of *Trifolium* both via the soil and by their interactions with other plant species. We measured significant increases in the availability of phosphates in soil sampled from containers containing worms (see also Bahl 1947; Lunt and Jacobson 1944; Aldag and Graff 1975; Nye 1955). Although soil analyses were only made at the end of the experiment to avoid disturbing the soil surface, it is likely that by the time the *Trifolium* begins to make a significant contribution to the species composition of the communities (after the dominance of both the *Senecio* and *Poa*) phosphate may well have been limiting in containers not containing earthworms. Consistent with this explanation, the growth of *Trifolium dubium* is significantly increased in either worm casts, or soil that previously supported a population

of earthworms, compared to growth rates in depleted soil from containers without earthworms (Fig. 5). Note, also, that the decaying bodies of dead worms may have influenced the nutrient balance of the containers. More work is required on this point.

Trifolium nodulation is also significantly enhanced in containers with earthworms (Fig. 3). The mechanism is unclear, although earthworms may concentrate rhizobium in the rhizosphere (Marsden and Alexander 1982). Nodulation may also be more effective in soils of low pH, but in this experiment the pH of soil containing earthworms ($\bar{x} = 6.43$) was not significantly lower ($P = 0.568$) than that sampled from eathworm-free treatments. If nitrogen is limited, which it may have been in all treatments, enhanced nodulation would place the *Trifolium* at a competitive advantage over non-leguminous species such as *Poa* and enhanced nodulation in the presence of worms should increase this advantage (cf. Stern and Donald 1962).

From day 135, much of the *Trifolium* was second-generation. Soil from different treatments had no direct effect on the germination of *Trifolium* seeds (Fig. 4). However real or false earthworm casts provided safe microsites for the establishment of *Trifolium* seedlings (Fig. 7) (see Fowler 1986; Harper et al. 1961; Harper 1977; Grubb 1977). *Trifolium dubium* germinates almost immediately, and in the Ecotron we observed large numbers of seedlings trapped in the *Poa* litter. These seedlings eventually shrivelled and died because their roots were unable to reach the soil surface, unless soil was artificially raised above the litter by the presence of real or false casts. Similar effects may explain the reduction in the competitiveness of *Poa* if litter is removed (see Bergelson 1990).

In summary, earthworms appear to benefit *Trifolium* by increasing nodulation and by creating safe microsites for establishment (by depositing casts). At the same time they increase concentrations of less mobile nutrients such as phosphorus, both by their action in mixing the soil, and possibly by increasing the rate of mobilisation via the burial of litter. There may also be unquantified effects of nutrients released from the decaying bodies of dead earthworms.

Although the activities of earthworms favour *Trifolium* directly in the short term, their long-term effects on community composition also appear to include the differential burial of seeds from different species: worms greatly increased the numbers of *Senecio* and *Poa* seeds buried in the seedbank, where they are no longer able to germinate and enter the community until brought to the surface by disturbance. The seedbank studies are consistent with previous observations that earthworms ingest seeds, and drag seeds below the soil surface (Darwin 1881; Kropac 1966). Earthworms are, however, selective in their effects, and prefer small grass seeds (Grant 1983). *Lumbricus terrestris*, one of the earthworm species used in the current experiments, prefers the seeds of *Poa annua* over the larger seeds of many *Trifolium* species (McRill and Sagar 1979).

When earthworms are combined with large snail populations, *Trifolium* loses its competitive advantage (days 111–161, Fig. 2d). These observations in a controlled environment demonstrate the importance of invertebrates in the dynamics of a simple plant community. Good evidence now exists that above-(Brown 1982, 1985, 1990; Brown et al. 1989; Brown and Gange 1988; Gange 1990; Mc Brien et al. 1983; Whittaker 1979) and below-ground (Brown and Gange 1989; Clements 1984; Clements et al. 1986; Brown and Gange 1990; Goldson and Proffitt 1985; Goldson et al. 1985) grazing by insect herbivores has a profound effect on the dynamics of simple ruderal communities. We observed that snails ate *Trifolium* preferentially, particularly the seedlings. From day 170 onwards, the depressing effect of snails on *Trifolium* was obvious, and *Trifolium* was eliminated from the snails-only treatment by the end of the experiment (Fig. 2d). Ammonia–nitrogen was significantly reduced in the presence of snails (Table 3). This may have been a function of the reduced proportion of nitrogen-fixing *Trifolium* in the community, together with a reduction in the availability of nitrogen, if it was bound up in the increased amount of *Poa* litter measured in snail treatments (Fig. 2c).

There were also interesting interactions between snails, worms and *Trifolium*; for example, individual snails from treatments containing worms were significantly larger, perhaps due to the increased productivity of the *Trifolium*.

Evidence for the role of molluscs in plant dynamics is substantial (Dirzo 1985; Edwards and Gillman 1987; Cottam 1986; Rees and Brown 1992), involving plant litter dynamics and effects on soil nutrients as well as herbivory. The present experiments suggest that earthworms could be of equal importance in the pattern of development of herbaceous plant communities, and that their effects interact with molluscs. In the Ecotron, earthworms and snails had strong effects on the abundance of *Trifolium*, by changing the availability of nutrients, germination microsites and the abundance of root nodules, and by altering the performance of potential competitors, by affecting litter and by worms burying seeds. All these factors have been hypothesised to be of major importance to plant community dynamics and composition (e.g. Harper et al. 1961; Harper 1977; Grubb 1977; Tilman 1982), and have shown to be affected by earthworms. They and the detrital food-chain in general cannot be ignored if we are to understand plant community dynamics.

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References

Aldag R, Graff O (1975) N-Fraktionen in Regenwurmlosung und deren Urpsprungsboden. Pedobiologia 15: 151–153

Bahl KN (1947) Excretion in the Oligochaeta. Biol Rev Cambridge Philos Soc 22: 109–147

Barley KP, Jennings AC (1959) Earthworms and soil fertility III The influence of earthworms on the availability of nitrogen. Aust J Agric Res 10: 364–370

Bergelson J (1990) Life after death: Site preemption by the remains of *Poa annua*. Ecology 71: 2157–2165

Brown VK (1982) The phytophagous insect community and its impact on early successional habitats. In: Visser JH, Minks AK (eds) Proceedings of the 5th International Symposium on Insect,

Plant relationships. Centre for Agricultural publishing and documentation, Wageningen. PUDOC, Wageningen, pp 205–212
Brown VK (1985) Insect herbivores and plant succession. Oikos 44: 17–22
Brown VK (1990) Insect herbivory and its effects on plant succession, In: Burdon JJ, Leather SR (eds) Pests, Pathogens and plant communities. Blackwell Scientific, Oxford, pp 42–62
Brown VK, Gange AC (1990) Insect herbivory below ground. Adv Ecol Res 20: 1–58
Brown VK, Gange AC, Gibson CWD (1988). Effects of insect herbivory on vegetation structure. In: Werger MJA, van der Aark PMJ, During HJ, Verhoeven JTA (eds) Plant form and vegetation structure. SPB Academic Publishing, The Hague, pp 263–279
Clements RO (1984) Control of insect pests in grassland. Span 27: 77–90
Clements RO, Bentley BR, Jackson CA (1986) The impact of granular formulations of phorate, terbufos, carbofuran, carbosulfan and thiofanox on newly sown italian ryegrass (*Lolium multiflorum*). Crop Protection 5: 389–394
Cottam DA (1986) The effects of slug grazing on *Trifolium repens* and *Dactylis glomerata* in monoculture and mixed swards. Oikos 47: 275–279
Darwin CR (1837) On the formation of mould. Trans Geol Soc Ser2. 5: 505–509
Darwin CR (1881) The formation of vegetable mould through the action of worms, with observations on their habits. Murray, London
Dirzo R (1985) The role of the grazing animal. In White J (ed) Studies on plant demography; a festschrift for John L Harper. Academic Press, New York, pp 343–355
Edwards RJ, Gillman MP (1987) Herbivores and plant succession. In: Crawley MJ, Edwards PJ, Gray A (eds) Colonisation, succession and stability. Blackwell Scientific, Oxford, pp 295–314
Fowler NL (1986) Microsite requirements for germination and establishment of three grass species. Am Midl Nat 115: 131–145
Gange AC (1990) Effects of insect herbivores on herbaceous plants. In: Burdon JJ, Leather SR (eds) Pests, pathogens and plant communities. Blackwell Scientific, Oxford, pp 42–62
Goldson SL, Proffitt JR (1985) Measurements of the impact of different larval densities of *Sitone discoides* Glylenhal (Coleoptera: Circulionidae) larvae and the associated patterns of adult infestation. NZ J Agric Res 26: 251–255
Goldson SL, Poured GW, Proffitt JR (1985) A study of the effects of *Sitone discoides* Glylenhal (Coleoptera: Circulionidae) larval feeding on the growth and development of lucerne (*Medicago sativa*). J Appl Ecol 24:153–161
Golterman HL Clymo RS, Ohnstad MAM (1978) Methods of physical and chemical analysis of freshwater. Blackwell Scientific, Oxford
Grant JD (1983) The activities of earthworms and the fates of seeds. In: Satchell JE (ed) Earthworm ecology. Chapman and Hall, London, pp 107–122
Grubb PJ (1977) The maintenance of species richness in plant communities: the importance of the regeneration niche. Biol Rev 82: 107–145
Harper JL (1977) The population biology of plants. Academic Press, London
Harper JL, Clatworthy JN, McNaughton IH, Sagar GR (1961) The evolution of closely related species living in the same area. Evolution 15: 209–227
Hoogerkamp M, Rogaar H, Eijsackers HJP (1983) Effect of earthworms on grassland on recently reclaimed polder soils in the Netherlands. In: Satchell JE (ed) Earthworm ecology. Chapman and Hall, London, pp 85–105
Hopp H, Slater CS (1948) Influence of earthworms on soil productivity. Soil Sci 66: 421–428
Jeanson C (1960) Evolution de la matier organique du sol sous l'action de *Lumbricus herculeus* CR Acad Sci 250: 3041–3043
Kropac Z (1966) Estimation of weed seeds in arable soil. Pedobiologia 6: 105–128
Lawton JH, Naeem S, Woodfin RM, Brown VK, Gange AC, Godfray HJC, Heads PA, Lawler S, Magda D, Thomas CD, Thompson L, Young S (in press) The Ecotron: A controlled environmental facility for the investigation of ecosystem processes. Philos Trans R Soc London B
Lee KE (1985) Earthworms, their ecology and relationships with soil and land use. Academic Press, Sidney.
Lunt HA, Jacobson GM (1944). The chemical composition of earthworm casts. Soil Sci 58: 367–375
Mackay AD, Syers JK, Gregg REH (1983) Plant availability of phosphorus in superphosphate and a phosphate rock as influenced by earthworms. Soil Biol Biochem 14: 281–287
Marsden EL, Alexander M (1982) Transport of *Rhizobium* and *Pseudomonas* through the soil. J Soil Soc Sci Am 46: 557–560
McBrien H, Harmsen R, Crowder A (1983). A case of insect grazing affecting plant succession. Ecology 64: 1035–1039
McRill RM, Sagar GR (1979) Earthworms and seeds. Nature 243: 482
Nye PH (1955) Some soil forming processes in the humid tropics IV. The action of the soil fauna. J Soil Sci 6: 73–83
Nydhal F (1976) Standard methods for the examination of water and wastewater. Talanta 23: 349–357
Parle JN (1963) A microbial study of earthworm casts. J Gen Microbiol 31: 13–22
Patton CJ, Crouch SR (1977) Methods for chemical analysis of water and wastes. Analytical Chemistry 49: 464–469
Rees M, Brown VK (1992) Interactions between invertebrate herbivores and plant competition. J Ecol 80: 353–360
Rhee JA van (1965) Earthworm activity and plant growth in artificial cultures. Plant Soil 22: 45–48
Satchell JE (ed) (1983) Earthworm Ecology: from Darwin to vermiculture. Chapman and Hall, London
Sears PD, Evans LT (1953) Pasture growth and soil fertility III. The influence of red and white clovers, superphosphate, lime and dung and urine on soil composition and on earthworm and grass-grub populations. NZ J Sci Tech 35: 42–52
Sharpley AN, Syers JK (1976) Potential role of earthworm casts for the phosphorus enrichment of run-off waters. Soil Biol Biochem 8: 341–346
Sharpley AN, Syers JK (1977) Seasonal variation in casting activity and in the amounts and release to solution of phosphorus forms in earthworm casts. Soil Biol Biochem 9: 227–231
Stern WR, Donald CM (1962) Light relationships in grass clover swards. Aust J Agric Res 13: 599–614
Stockdill SMJ (1966) The effects of earthworms of pastures. Proc NZ Ecol Soc 13: 68–75
Stockdill SMJ (1982) Effects of introduced earthworms on the producitivity of New Zealand pastures. Pedobiologia 24: 29–35
Tilman D (1982) Resource competition and community structure. Princeton University Press, Princeton
Tisdall JM (1978) Ecology of earthworms in irrigated orchards. In: Emerson WW, Bond RD, Dexter AR (eds) Modification of soil structure. Wiley, Chichester, pp 297–303
Whittaker JB (1979) Invertebrate grazing, competition and plant dynamics In: Anderson RM, Turner BD, Taylor LR (eds) Population dynamics (Symp Br Ecol Soc 20). Blackwell Scientific, Oxford, pp 207–222