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Carbon dynamics in a model grassland with functionally different soil communities

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Summary

1. Impacts of environmental changes on carbon cycling in terrestrial ecosystems are typically correlated with shifts in the composition, biomass and activity of soil faunal communities. Given the role of soil fauna in decomposition, shifts in soil faunal communities may further modify the carbon dynamics of a perturbed system.

2. To test this possibility, we manipulated soil community composition in model grassland ecosystems and used a $^{13}CO_2$-tracer to follow the assimilation, retention and pathways of recent, photosynthetically fixed carbon. The community treatments formed a nested hierarchy of functional complexity: (1) microbiota only; (2) microbiota and mesofauna; (3) microbiota, mesofauna and macrofauna.

3. These treatments significantly affected the rate of decline in ^{13}C-label respiration rate and the amount of ^{13}C-label retained by the communities: while the rate of decline in ^{13}C-label respiration rate was greatest in the microbiota treatment, the treatment with both micro- and mesofauna retained less ^{13}C-label than either the more functionally complex or simple treatments. The presence of macrofauna altered the utilization of ^{13}C-label by Collembola and Enchytraeidae: they decreased the mass of ^{13}C-label utilized by Collembola and increased that utilized by Enchytraeidae.

4. Our results suggest that soil community composition may play a key role in regulating the dynamics of recent, photosynthetically fixed carbon.

Key-words: ^{13}C, carbon dynamics, decomposition, functional diversity, rhizodeposition, soil fauna

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Introduction

Environmental change, such as increasing concentrations of atmospheric CO_2, nitrogen deposition and land-use change, may disrupt the dynamics of carbon flux in terrestrial ecosystems (Schimel *et al.* 2001; Schlesinger & Lichter 2001; Neff *et al.* 2002). These impacts on ecosystem carbon (C) dynamics are typically associated with changes in the composition of soil faunal communities (Jones *et al.* 1998; Wardle *et al.* 1998; Wolters *et al.* 2000). Given the important functional role of soil fauna in decomposition and mineralization of dead organic matter (Swift *et al.* 1979; Petersen & Luxton 1982), alterations in soil faunal community composition may further modify the C dynamics of a system perturbed by environmental change.

When larger-bodied soil fauna are excluded from surface litter, mass loss from this litter is reduced

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(Bradford *et al*. 2002a). Such exclusions have been shown to alter plant litter quality and litter species-richness effects on litter decomposition rates (Smith & Bradford 2003; Hättenschwiler & Gasser 2005). The effects of manipulating which soil fauna are present/absent on C-cycling processes such as plant and net ecosystem productivity are less conclusive (Wardle *et al*. 2004). For example, while positive (Laakso & Setälä 1999; Liiri *et al*. 2002a) and negative (Bardgett & Chan 1999; Liiri *et al*. 2002b) effects of faunal species/groups on plant growth have been demonstrated, such effects have been cancelled out when additional faunal groups have been included in experimental systems (Alphei *et al*. 1996). These findings, and those from other studies (e.g. Hunt & Wall 2000; Bradford *et al*. 2002a; Cole *et al*. 2004), where neutral effects of soil faunal group/species removal on plant and net ecosystem productivity have been demonstrated, are reinforcing the assumption that functional redundancy is a common feature in soils (Andrén & Balandreau 1999; Laakso & Setälä 1999; Setälä, Berg & Jones 2005).

Contrary to the assumption of functional redundancy in soils, De Deyn *et al*. (2003) demonstrated that differences in plant productivity arise when communities are exposed to soil fauna derived from grasslands at different successional stages. In addition, both positive and negative effects on soil respiration were observed by Heemsbergen *et al*. (2004) when experimental microcosms contained different combinations of earthworms and soil macro-arthropods. Together, these studies suggest that soil faunal community composition is a potentially important determinant of ecosystem C dynamics. One aspect of these C dynamics, overlooked until recently (Johnson *et al*. 2005) from the context of soil community composition, is the fate of recent photosynthetically fixed C. Flux of this C into decomposer food webs, through processes such as rhizodeposition, may fuel mineralization pathways that make a substantial contribution to ecosystem C exchange (van Hees *et al*. 2005).

Here, soil faunal community composition is manipulated by establishing a series of microcosms with a nested hierarchy of organism body widths (e.g. Setälä *et al*. 1996). This provides a treatment gradient of increasing functional complexity, because body width provides a functional classification (Beare *et al*. 1995; Brussaard *et al*. 1997) where a single trait (size) is expected to affect many ecosystem processes and pools (see Hooper *et al*. 2005). Body width provides a good functional classification because: (a) it correlates with metabolic rate, generation time, population density and food size (Peters 1983); (b) the physical structure of the soil habitat constrains access to resources for certain body widths and hence modulates interactions between organisms (Brussaard *et al*. 1997); and (c) to some extent, body width maps onto the three hierarchical levels of interaction (microfood webs, litter transformers and ecosystem engineers) in soil food webs where, with increasing body width of fauna, the relationship of fauna with microflora shifts from predation to mutualism (Lavelle 1997). In addition, given the limited understanding of soil food-web interactions (Wardle 2002), body width provides an experimentally tractable manipulation. As a manipulation it has both fundamental (for the reasons stated above) and applied scientific relevance because, in the context of applied relevance, human activities such as land-use conversion to crop land have a disproportionately negative effect on larger-bodied fauna (Wardle 2002).

We combine our microcosm approach with ^{13}C-tracer techniques to investigate whether the functional complexity of soil faunal communities might influence the assimilation, retention and pathways of recent, photosynthetically fixed C. Earlier work on these experimental communities showed that although processes key to C cycling, such as litter decomposition, responded markedly to altered soil faunal community composition (Bradford *et al*. 2002b), measures of total ecosystem C balance (such as net ecosystem productivity) were insensitive to the manipulations (Bradford *et al*. 2002a). The non-isotopic measures employed in these two studies (Bradford *et al*. 2002a; 2002b) did not, however, allow us to elucidate how the soil community treatments might affect the fate of recent, photosynthetically fixed C.

We test two hypotheses relating to the fate of recently fixed C. The first is generated from the expectation that soil mesofauna and macrofauna facilitate the decomposition of plant C inputs to soil through processes such as fragmentation, herbivory, microbivory and soil engineering. Together, these processes serve to stimulate the activity of, and maintain the availability of C substrates to, soil micro-organisms (Lussenhop 1992; Gange 2000; Hedlund & Öhrn 2000). Hence we hypothesize that the rate of decline in ^{13}C-label respiration rate will be greater (steeper slope) with decreasing functional complexity (experimental removal of larger fauna) (H1). Despite the fact that larger fauna are expected to stimulate micro-organisms in the shorter term, they create decomposition products that are, in the longer term, more resistant to microbial mineralization (Petersen & Luxton 1982; Verhoef & Brussaard 1990). Our second hypothesis is based on this stabilization of C inputs. We hypothesize that experimental removal of larger fauna will decrease the amount of ^{13}C-label retained within the experimental communities (H2). Thus our expectations are that the net effect of soil mesofauna and macrofauna will be to make plant C inputs more consistently available to microbial mineralization in the short term (H1); but in the longer term will increase their retention time (H2). If this occurs, we would expect to observe the greatest ^{13}C-label mineralization rates from the functionally simplest experimental communities in the short term, but the lowest rates in the longer term.

Materials and methods

MICROCOSM ESTABLISHMENT AND TREATMENT DESIGN

Fifteen terrestrial microcosms (1 m^2 surface area) were established and maintained in the Ecotron controlled environment facility (Lawton 1996) for 312 days. The microcosms were constructed from grassland at Sourhope Experimental Farm (UK National Grid Reference NT855196) in Scotland. Soils from the site were collected, roughly homogenized by horizon, then treated with methyl bromide (CH_3Br) to remove the seed bank, mesofauna and macrofauna. The soil profile was then reconstructed (to a depth of 303 mm) by horizon; the upper mineral horizon was split into two layers because the upper portion had greater organic matter content. Horizons and their depths were as follows: FH (35 mm), H (20 mm), Ah 'upper' (69 mm), Ah 'lower' (114 mm), AB (65 mm).

Seeds of the 10 most dominant plants at the Sourhope grassland were collected, glasshouse-germinated and introduced into the microcosms as seedlings in a randomly generated grid pattern that was repeated for all microcosm communities. In total, 384 seedlings were introduced into each community and were, in the same order of decreasing dominance as at the field site: *Agrostis capillaris* L., *Festuca rubra* L., *Nardus stricta* L., *Anthoxanthum odoratum* L., *Poa pratensis* L., *Trifolium repens* L., *Holcus mollis* L., *Potentilla erecta* L., *Galium saxatile* L. and *Luzula multiflora* (Ehrh.) Lej. All species established and persisted.

Each microcosm was housed within a $2 \times 2 \times 2$-m Ecotron walk-in chamber. The Ecotron facility permits tight environmental control: simulated abiotic conditions in each chamber are effectively identical (Lawton 1996). Photoperiod was 18 h, including a gradual dawn and dusk of 2 h each. Temperature and relative humidity followed sine curves between 21·1 °C during the day and 9·5 °C at night, and 83% after rainfall (3·5 mm day^{-1}) to a minimum of 63%, respectively.

Soil biota were introduced selectively to create communities that differed in organism body width. If the adult was not soil-dwelling, organisms were assigned to a size class based on their largest juvenile stage. We adhered to the general size classification for terrestrial decomposer food webs (Swift *et al.* 1979), establishing three treatments that formed a gradient of increasing functional complexity: (1) microbiota only (<100 µm diameter; primarily bacteria, fungi, Protozoa, Nematoda); (2) microbiota and mesofauna (100 µm to 2 mm diameter; primarily Collembola, Acari, Enchytraeidae); (3) microbiota, mesofauna and macrofauna (>2 mm diameter; primarily earthworms, slugs, insect larvae, staphylinid beetles). Hereafter, these treatments are referred to as microbiota, mesofauna and macrofauna treatments, respectively.

Soils were repeatedly inoculated with microbial solutions, derived using various physical extraction techniques (Bradford *et al.* 2002a) from Sourhope grassland turves. Microfauna, mesofauna and the majority of the macrofauna were also extracted from Sourhope turves using wet- (Nematoda and Enchytraeidae) or dry- (other mesofauna and macrofauna) funnel techniques. Two earthworm species (*Allolobophora chlorotica* Sav. and *Lumbricus rubellus* Hoffm.) and a mollusc (*Arion ater* L.) were obtained from biological suppliers (Blades Biological, Edenbridge, UK). These macrofauna species were present at the Sourhope grassland, but to obtain them in sufficient biomass to establish our experiment was deemed too destructive to the site.

Treatments were randomly assigned within five blocks defined by microcosm planting order. The block structure was used in all further set-up and in sampling, such that all procedures were conducted, in a random order, by block. Microcosms were constructed over 7 months: day 1 was assigned at the end of this period, the day after the last faunal inoculation was made. Microcosms were established gradually to permit adequate time for development of a 'mature' grassland plant community, as well as growth of the microflora and faunal communities. This timeline also permitted diminishing of the C and nutrient pulse, inherent to microcosm construction, prior to the experiment. Biomass and numbers of soil microflora and fauna once the microcosms were established (designated day 1) and on day 257 (the time of ^{13}C-labelling and thus the start of the experimental work presented here) are provided by Bradford *et al.* (2002a), as are further details of experimental set-up.

^{13}C-LABELLING

We followed the movement of a ^{13}C-label into plant foliar material, then through the soil community and back to the atmosphere as respired CO_2 across a 52-day period. Communities were ^{13}C-labelled using a mobile laboratory that administers a controlled flow of $^{13}CO_2$ at ambient CO_2 concentrations (≈ 370 µl l^{-1}). This stable isotope-delivery system is described by Ostle *et al.* (2000). With this system, six microcosm communities could be labelled at one time. Two experimental blocks were pulse-labelled on day 255 and two blocks on day 257. Sampling of these blocks was staggered appropriately. The non-labelled block provided the natural abundance values for the measured C pools (see below).

To introduce the ^{13}C-label, microcosm communities were capped with a chamber constructed from photosynthetically active radiation-transparent polyester sheeting (Mylar). Labelling continued for 6 h during the high-light (536 µmol m^{-2}) period of the diurnal cycle. The rapid turnover time of air in the chambers meant that dilution of the $^{13}CO_2$ by community respiration was minimal.

FOLIAR, SOIL COMMUNITY AND CO_2 ANALYSES FOR ^{13}C-LABEL CONTENT

Clippings of plant foliar material were taken immediately prior to and immediately after ^{13}C-labelling, and were used to estimate the amount of ^{13}C-label assimilated by the plant community. To generate a representative community sample, the proportion of dry foliar biomass of each species to include was quantitatively based on plant species-abundance data, and within each microcosm at least five individuals of each plant species were sampled. Clipped material was dried at 40 °C and then milled under liquid N_2.

Micro-arthropods (Collembola and Acari) were dry-extracted from 50 mm diameter organic horizon cores (55 mm depth) using Tullgren funnels. Enchytraeidae were wet-extracted from cores of the same size. At 2 days after the pulse, three such cores were sampled per microcosm for the micro-arthropods and three for the Enchytraeidae. Separate but single cores of the same size were used for extraction of Nematoda and determination of microbial biomass C. At 8 and 18 days after the pulse, the micro-arthropods and Enchytraeidae were extracted from three cores in total to minimize soil disturbance. Each core was halved longitudinally, with one half used for micro-arthropods and one for Enchytraeidae. This still provided a sampling area greater than recommended for extracting these fauna (Coleman *et al.* 2004).

Micro-arthropods were collected into salt water to kill the animals and prevent predation. Extractions continued across 48 h, but collected fauna were sorted and dried at 40 °C after each 24-h period to avoid biomass decay. Dry samples were weighed and milled under liquid N_2. Enchytraeidae were freeze-dried prior to weighing. Nematoda were also freeze-dried, after extraction using a tray technique (Whitehead & Hemming 1965). Microbial biomass C was estimated using modified chloroform fumigation extraction (Vance *et al.* 1987), and provided for isotope analyses as potassium sulphate (K_2SO_4) extracts. All fauna were analysed whole (including gut contents). It was deemed too destructive to the communities to sample macrofauna and roots.

Community-respired CO_2 samples were collected 2, 5, 8, 12, 18, 32 and 52 days post-labelling using a static cover-box technique (Holland *et al.* 1999). Gas samples from the cover-box headspaces were stored in 12 ml Exetainer tubes (Labco Ltd, High Wgcombe, UK) for <72 h prior to analysis.

All samples were analysed for C concentration and $^{13}C/^{12}C$ ratios by continuous-flow isotope-ratio mass spectrometry (IRMS) at the NERC ^{15}N Stable Isotope Facility. Plant and soil biota samples were analysed using an elemental analyser (Carlo Erba/Fisons, Milan, Italy) linked to a modified IRMS (Dennis Leigh, Keele, UK) and CO_2 samples using a trace gas preconcentration unit coupled to an Isoprime IRMS (Micromass, Manchester, UK). Analytical precision was ±0·1 $\delta^{13}C‰$. Internal gas standards and solid reference materials were calibrated to PDB (Pee Dee Belemnite) (Craig 1957).

STATISTICAL ANALYSES

To determine how the treatments affected assimilation and retention of the ^{13}C-label, ANOVA was used. Block was included as a non-interacting factor. To assess how the treatments affected the rate of change in ^{13}C-label respiration, we used linear mixed-effects modelling. For this approach, fixed effects were time (continuous variable) and soil community treatment (categorical variable). To identify the unit of repeated measurement across time, the random effects were defined as chamber nested within block.

The linear mixed-effects model structure was used to determine how the treatments affected the utilization of the ^{13}C-label by the microbial biomass C and Collembola pools. This model structure was unable to cope with missing values for the Nematoda, Enchytraeidae and Acari pools. Given that (as with the Collembola) there were no apparent time effects in these data, we dropped time from the model structure and analysed these pools using mean values across time for each replicate (either the mean value across all three time points, or only two time points when there was a missing value). All data were natural log-transformed prior to analysis. We follow the convention of reporting significance assuming $\alpha < 0{\cdot}05$. We report significance at one of three levels: $P < 0{\cdot}05$, 0·01 or 0·001; for marginally significant ($P < 0{\cdot}1$ but $\geq 0{\cdot}05$) results we report the actual P value. Following convention, we report non-significant effects as $P > 0{\cdot}05$, but note that in all such instances the actual P value was $\geq 0{\cdot}1$.

Treatment effects on utilization of ^{13}C-label by the biota pools can be assessed in three different ways, each defined by how the ^{13}C-label values for a pool are expressed. The ^{13}C-label mass values can be expressed (i) in terms of absolute mass of ^{13}C-label; (ii) as their absolute mass relative to the ^{13}C-label mass assimilated by the plant community; and (iii) as their absolute mass relative to the total mass of C in a biota pool. The second expression (mass relative to that assimilated by the plant community) gives information similar to the first expression (absolute mass), except that differences in ^{13}C-label assimilation are corrected for. The first expression is relevant because, in contrast to experiments where a set amount of ^{13}C-label is applied, the community treatments can influence the absolute uptake of the ^{13}C-label. The third expression (relative to the total mass of C in a biota pool) permits assessment of whether the amount of C acquired by the pool that is derived from recent photosynthate, relative to that acquired from other substrates, is affected.

Results

The ^{13}C-labelling resulted in enrichments of ^{13}C concentration of the plant foliar material, which ranged

between 540 and 850‰. There was no significant treatment effect on the absolute mass of ^{13}C-label assimilated into plant community foliar material ($F_{2,5} = 0{\cdot}48$, $P > 0{\cdot}05$), or the mass assimilated as a proportion of the plant community biomass ($F_{2,5} = 0{\cdot}49$, $P > 0{\cdot}05$). There was no ^{13}C-contamination of the 'natural abundance' communities.

The rate of change (slope) in ^{13}C-label respiration rate for each treatment initially declined rapidly and then began to plateau (Fig. 1). The slope was significantly greater for the microbiota treatment than for the other two treatment communities (time × community interaction: $F_{2,69} = 11$, $P < 0{\cdot}001$; Fig. 1).

There was no significant treatment effect on the absolute mass of ^{13}C-label respired from the communities ($F_{2,5} = 1{\cdot}1$, $P > 0{\cdot}05$), although the mean for the mesofauna treatment was greater than for the other two treatments: mean ± 1 SE values for the microbiota, mesofauna and macrofauna treatments were 26 ± 16·1, 46 ± 14·1 and 25 ± 11·1 (mg ^{13}C-label m^{-2}), respectively. When expressed as a proportion of the mass of ^{13}C-label assimilated (which accounts for differences between replicates in ^{13}C-label assimilation), loss of the label was significantly greater ($F_{2,5} = 6{\cdot}2$, $P < 0{\cdot}05$) in the mesofauna treatment (Fig. 2).

The absolute and relative masses of ^{13}C-label in the microbial biomass C pool declined significantly ($P < 0{\cdot}001$) with time (Table 1) but were not significantly affected by treatment ($P > 0{\cdot}05$ for the community and time × community effects). In contrast, there were significant ($P < 0{\cdot}05$) or marginally significant ($P < 0{\cdot}1$) treatment effects on the fauna. For the Enchytraeidae pool, the absolute and relative mass of ^{13}C-label was greatest ($F_{1,6} = 5{\cdot}67$, $P = 0{\cdot}055$; $F_{1,6} = 6{\cdot}03$, $P < 0{\cdot}05$, respectively) in the macrofauna treatment (Table 1). For the Collembola pool the mass of ^{13}C-label, when expressed relative to that assimilated by the plants, was greatest ($F_{1,3} = 8{\cdot}52$, $P = 0{\cdot}062$) in the mesofauna treatment (Table 1). There were no significant ($P > 0{\cdot}05$) treatment effects on the Nematoda or Acari pools (Table 1).

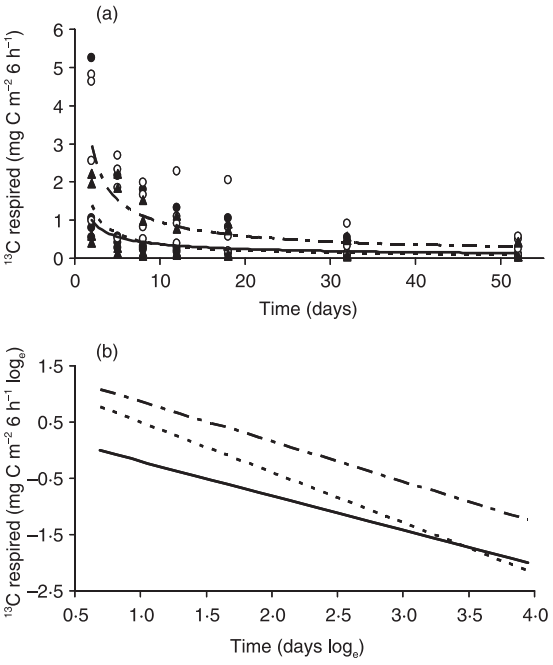

Fig. 1. Rate of change in ^{13}C-label respiration rate across the 52-day measurement period in the microbiota (dotted line, ●), mesofauna (hatched line, ○) and macrofauna treatments (solid line, ▲). The slope was significantly greater in the microbiota treatment ($P < 0{\cdot}001$). The relationship between ^{13}C-label respiration rate and time was best described by a power function; for data from any one replicate, these power functions explained at minimum 81% and maximum 98% of the variation. Given the significant effect of block (which was related to large differences between replicates of the same treatment across blocks), when regression lines were fitted to data from a community (as opposed to a replicate), then much less variation across time was explained. That is, the fits (r^2 values) for the microbiota, mesofauna and macrofauna treatments were 0·55, 0·53 and 0·25, respectively. (a) Untransformed data; (b) transformed data. Transformation to linearize the relationship was carried out by taking the natural log of both dependent and independent data. For clarity, in (b) only mean regression lines are shown. Those regression lines shown are fitted using all data from a specific community treatment.

Discussion

Our first hypothesis was that the rate of decline in ^{13}C-label respiration rate would be greater (steeper slope) with decreasing functional complexity (removal of larger fauna) because mesofauna and macrofauna are expected to maintain the supply of mineralizable C to the microbial biomass (Lussenhop 1992; Gange 2000; Hedlund & Öhrn 2000). Our hypothesis was partially supported by our data: the rate of decline in ^{13}C-label respiration rate was significantly greater when meso- and macrofauna were absent from the communities (Fig. 1). When mesofauna were present without macrofauna, the dynamics were the same as when macrofauna were also present (Fig. 1). This suggests that mesofauna may compensate for the absence of macrofauna in terms of maintaining the supply across time of recent, photosynthetically fixed C for

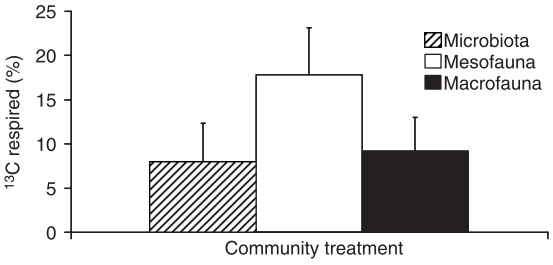

Fig. 2. Effects of soil community functional complexity on ^{13}C-label retention. Values are ^{13}C-label mass respired (mean ± 1 SE; $n = 4$) as a percentage of that assimilated. The mesofauna treatment retained significantly less ($P < 0{\cdot}05$) label than either the microbiota or macrofauna treatments.

Table 1. Mass of ^{13}C-label in soil biota pools expressed (a) in absolute terms; (b) relative to label mass assimilated by the plant community; (c) relative to total mass of carbon in the respective biota pool

		Treatment communities		
Pools		Microbiota	Mesofauna	Macrofauna
(a)	Day	(mg C m^{-2})	(mg C m^{-2})	(mg C m^{-2})
Microbial	2	22·5 ± 3·59	19·4 ± 3·20	15·2 ± 1·40
	8	11·0 ± 1·80	9·1 ± 3·25	8·8 ± 1·13
	18	6·2 ± 0·50	5·3 ± 1·31	5·0 ± 0·48
Nematoda	na	$2{\cdot}0 \times 10^{-2} \pm 1{\cdot}02 \times 10^{-2}$	$1{\cdot}4 \times 10^{-2} \pm 4{\cdot}86 \times 10^{-3}$	$2{\cdot}5 \times 10^{-2} \pm 1{\cdot}47 \times 10^{-2}$
Collembola	na	na	$3{\cdot}6 \times 10^{-1} \pm 2{\cdot}79 \times 10^{-1}$	$5{\cdot}2 \times 10^{-2} \pm 8{\cdot}61 \times 10^{-3}$
Acari	na	na	$5{\cdot}7 \times 10^{-2} \pm 4{\cdot}17 \times 10^{-2}$	$2{\cdot}1 \times 10^{-2} \pm 8{\cdot}79 \times 10^{-3}$
Enchytraeidae	na	na	$2{\cdot}9 \times 10^{-3} \pm 1{\cdot}29 \times 10^{-3}$	$1{\cdot}2 \times 10^{-2} \pm 3{\cdot}65 \times 10^{-3}$
(b)	Day	(%)	(%)	(%)
Microbial	2	7·2 ± 1·11	8·5 ± 2·80	6·5 ± 0·80
	8	3·9 ± 1·45	5·0 ± 2·76	3·7 ± 0·34
	18	2·1 ± 0·49	2·6 ± 1·17	2·1 ± 0·31
Nematoda	na	$6{\cdot}0 \times 10^{-3} \pm 2{\cdot}79 \times 10^{-3}$	$7{\cdot}3 \times 10^{-3} \pm 3{\cdot}46 \times 10^{-3}$	$1{\cdot}3 \times 10^{-2} \pm 8{\cdot}76 \times 10^{-3}$
Collembola	na	na	$8{\cdot}3 \times 10^{-2} \pm 4{\cdot}81 \times 10^{-2}$	$2{\cdot}3 \times 10^{-2} \pm 5{\cdot}33 \times 10^{-3}$
Acari	na	na	$1{\cdot}4 \times 10^{-2} \pm 7{\cdot}71 \times 10^{-3}$	$1{\cdot}0 \times 10^{-2} \pm 5{\cdot}41 \times 10^{-3}$
Enchytraeidae	na	na	$1{\cdot}2 \times 10^{-3} \pm 5{\cdot}47 \times 10^{-4}$	$5{\cdot}5 \times 10^{-3} \pm 2{\cdot}06 \times 10^{-3}$
(c)	Day	(%)	(%)	(%)
Microbial	2	$4{\cdot}8 \times 10^{-2} \pm 5{\cdot}80 \times 10^{-3}$	$4{\cdot}3 \times 10^{-2} \pm 7{\cdot}09 \times 10^{-3}$	$4{\cdot}3 \times 10^{-2} \pm 5{\cdot}47 \times 10^{-3}$
	8	$2{\cdot}3 \times 10^{-2} \pm 1{\cdot}81 \times 10^{-3}$	$1{\cdot}9 \times 10^{-2} \pm 6{\cdot}07 \times 10^{-3}$	$2{\cdot}5 \times 10^{-2} \pm 3{\cdot}45 \times 10^{-3}$
	18	$1{\cdot}3 \times 10^{-2} \pm 7{\cdot}96 \times 10^{-4}$	$1{\cdot}1 \times 10^{-2} \pm 2{\cdot}49 \times 10^{-3}$	$1{\cdot}4 \times 10^{-2} \pm 1{\cdot}19 \times 10^{-3}$
Nematoda	na	$1{\cdot}8 \times 10^{-2} \pm 6{\cdot}36 \times 10^{-3}$	$1{\cdot}8 \times 10^{-2} \pm 7{\cdot}46 \times 10^{-3}$	$3{\cdot}9 \times 10^{-2} \pm 2{\cdot}17 \times 10^{-2}$
Collembola	na	na	$8{\cdot}4 \times 10^{-2} \pm 2{\cdot}08 \times 10^{-2}$	$8{\cdot}9 \times 10^{-2} \pm 1{\cdot}49 \times 10^{-2}$
Acari	na	na	$7{\cdot}4 \times 10^{-2} \pm 4{\cdot}80 \times 10^{-2}$	$3{\cdot}4 \times 10^{-2} \pm 1{\cdot}02 \times 10^{-2}$
Enchytraeidae	na	na	$5{\cdot}9 \times 10^{-3} \pm 1{\cdot}00 \times 10^{-3}$	$8{\cdot}2 \times 10^{-3} \pm 1{\cdot}77 \times 10^{-3}$

Values are means ± 1 SE (n = 4). Given a significant ($P < 0{\cdot}001$) effect of time, those values for the microbial biomass C are given for each measurement day; there were no time effects for any of the fauna pools, so values presented are means of the mean value across time for each replicate. na, Not applicable.

mineralization to CO_2. However, in the context of this functional parameter, the microbiota themselves cannot compensate for the absence of both meso- and macrofauna.

Our second hypothesis, that there would be greater ^{13}C-label retention with greater functional complexity, was not supported by the data: relative retention of the ^{13}C-label was less in the mesofauna treatment and did not differ significantly between the most functionally simple or complex treatments (Fig. 2). Thus the effect of the presence of mesofauna on ^{13}C-label retention was negated when macrofauna were also present. This finding adds to those studies (Alphei *et al.* 1996; Heemsbergen *et al.* 2004) which report that additional soil faunal groups may cancel out effects of other fauna on ecosystem processes. In addition, our finding (loss of macrofauna decreased retention of the ^{13}C-label by ≈ 10% when mesofauna were present; Fig. 2) may have implications for C sequestration. Given that we followed mineralization of the ^{13}C-label across only 52 days, further work across longer time-scales measuring changes in soil organic C fractions is required to establish the potential significance of our finding for C storage.

Within the soil communities, the only fauna pools to be affected by treatment were the Collembola and Enchytraeidae. The greater ^{13}C-label mass in the Collembola, in the absence of macrofauna, was related to the greater biomass of this pool in the mesofauna than the macrofauna treatment (Table 2). The same relationship held for the Enchytraeidae; except for this pool, the ^{13}C-label mass and Enchytraeidae mass were greater in the macrofauna treatment. Notably, the amount of ^{13}C-label acquired relative to the total mass of C in these two faunal pools was unaffected by treatment. This may suggest that the pools' trophic group and/or diet composition was unaffected by macrofauna presence/absence. This may have occurred either because macrofauna do not alter the species composition of these pools or, if composition is affected, because those species increasing in dominance have the same feeding ecology as those they are replacing. Whether resource competition and facilitation with macrofauna explain the lower Collembola and greater Enchytraeidae label mass, respectively, cannot be elucidated from our study.

Both our hypotheses relating to the ^{13}C-label dynamics under the different soil communities were developed

Table 2. Biomass of soil biota pools

Biota pool	Biomass (mg m^{-2}) in treatment communities		
	Microbiota	Mesofauna	Macrofauna
Microbial (carbon)	$465 \times 10^2 \pm 396{\cdot}8 \times 10^1$	$453 \times 10^2 \pm 231{\cdot}1 \times 10^1$	$359 \times 10^2 \pm 223{\cdot}3 \times 10^1$
Nematoda	189 ± 40·8	196 ± 21·7	146 ± 17·0
Collembola	0	770 ± 477·7	156 ± 36·0
Acari	0	134 ± 39·9	138 ± 24·1
Enchytraeidae	0	156 ± 94·2	357 ± 90·9

Values are means ± 1 SE (n = 4).

from the expectation of how mesofauna and macrofauna directly and indirectly affect heterotrophic mineralization of plant C inputs to soils. However, we recognize that observed changes in plant community composition and mycorrhizal abundance (Bradford *et al*. 2002a) under the different soil community treatments might also have played a role, as well as other potential mechanisms. For example, Johnson *et al*. (2005) demonstrated that Collembola alter the dynamics of recent, photosynthetically fixed C flux through the mycorrhizosphere. Our work cannot therefore pinpoint any one specific mechanistic scenario to explain the ^{13}C-label dynamics we observed; further work is required to understand better soil community impacts on the dynamics of recent, photosynthate C. Such mechanistic insights are also limited because of our nested experimental design where the more complex treatments, in addition to larger-bodied fauna, contained those fauna and microbes in simpler communities. Thus we cannot ascertain whether treatment effects resulted from inclusion of larger-bodied fauna *per se*, or whether they arose because the larger-bodied fauna altered the abundance, biomass or composition of those microbes or fauna also present in the simpler communities. Finally, our results must be seen in the context of artificially assembled communities (Lawton 1996) and thus caution should be exercised when extrapolating findings.

Our results demonstrate that varying soil faunal community complexity has the potential to affect the respiration, retention and movement into soil fauna pools of recently, photosynthetically fixed C. Our results further suggest that fauna of smaller body widths do not necessarily compensate functionally for the absence of larger-bodied fauna. In the context of environmental changes that disproportionately negatively affect larger-bodied fauna (e.g. conversion of land use to crop land; Wardle 2002), we might thus attribute changes in ecosystem C dynamics, at least in part, to shifts in soil faunal community composition. Given that rhizodeposited C may constitute the dominant flux of plant C to soils (van Hees *et al*. 2005), understanding interactions between soil faunal community composition and the dynamics of recent, photosynthetically fixed C should be made a research priority.

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