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Influence of warming and enchytraeid activities on soil CO_2 and CH_4 fluxes

M.J.I. Briones[a,b,*], J. Poskitt[b], N. Ostle[b]

[a]*Departamento de Ecología y Biología Animal, Facultad de Biologia, Universidad de Vigo, 36310 Vigo, Spain*
[b]*Centre for Ecology and Hydrology, Lancaster Environment Centre, Library Avenue, Bailrig, Lancaster, LA1 4AP, UK*

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Abstract

To determine the sum of 'direct' and 'indirect' effects of climatic change on enchytraeid activity and C fluxes from an organic soil we assessed the influence of temperature (4, 10 and 15 °C incubations) on enchytraeid populations and soil CO_2 and CH_4 fluxes over 116 days. Moisture was maintained at 60% of soil dry weight during the experimental period and measurements of enchytraeid biomass and numbers, and CO_2 and CH_4 fluxes were made after 3, 16, 33, 44, 65, 86 and 116 days. Enchytraeid population numbers and biomass increased in all temperature treatments with the greatest increase produced at 15 °C (to over threefold initial values by day 86). Results also showed that enchytraeid activity increased CO_2 fluxes by 10.7 ± 4.5, 3.4 ± 4.0 and 26.8 ± 2.6% in 4, 10 and 15 °C treatments, respectively, with the greatest CO_2 production observed at 15 °C for the entire 116 day incubation period ($P < 0.05$). The soil respiratory quotient analyses at lower temperatures (i.e. 4–10 °C) gave a Q_{10} of 1.7 and 1.9 with and without enchytraeids, respectively. At temperatures above 10 °C (i.e. 10–15 °C) Q_{10} significantly increased ($P < 0.01$) and was 25% greater in the presence of enchytraeids ($Q_{10} = 3.4$) than without ($Q_{10} = 2.6$). In contrast to CO_2 production, no significant relationships were observed between net CH_4 fluxes and temperature and only time showed a significant effect on CH_4 production ($P < 0.01$).

Total soil CO_2 production was positively linked with enchytraeid biomass and mean soil CO_2–C production was 77.01 ± 6.05 CO_2–C μg mg enchytraeid tissue^{-1} day^{-1} irrespective of temperature treatment. This positive relationship was used to build a two step regression model to estimate the effects of temperature on enchytraeid biomass and soil CO_2 respiration in the field. Predictions of potential CO_2 production were made using enchytraeid biomass data obtained in the field from two upland grassland sites (Sourhope and Great Dun Fell at the Moor House Nature Reserve, both in the UK). The findings of this work suggest that a 5 °C increase in atmospheric temperature above mean ambient temperature could have the potential to produce a significant increase in enchytraeid biomass resulting in a near twofold increase in soil CO_2 release from both soil types. The interaction between temperature and soil biology will clearly be an important determinant of soil respiration responses to global warming.

Keywords: Climate change; Enchytraeids; Greenhouse gas; Soil respiration; Q_{10}

1. Introduction

The nature of the coupling between climate and the terrestrial C cycle will need to be elucidated to predict future responses of ecosystems to global change (IPCC). However, our understanding of the biological mechanisms involved in the regulation of soil C release to the atmosphere remains limited. One of the major uncertainties in current GCM predictions is the response of soil respiration to changes in atmospheric temperature (Luo et al., 2001). This information is essential to model the potential effects of changes in the biotic production of greenhouse gases (CO_2 and CH_4) on atmospheric temperature and the feed-back effects of the resultant climatic change on subsequent soil C release (Cox et al., 2000).

Recent findings suggest that the C storage potential of grassland soil has already been attained and that the ability of soils to continue as C sinks is limited (Gill et al., 2002). The preservation of existing soil C stocks will therefore

* Corresponding author. Tel.: +34-986-812584; fax: +34-986-812556.
E-mail address: mbriones@uvigo.es (M.J.I. Briones).

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depend upon the contribution of soil heterotroph activity to C release, particularly at the interface between live plant roots and the soil in the rhizosphere (Paterson et al., 1997). Increased activity of the rhizosphere heterotroph community, that includes soil microorganisms and invertebrates, as a result of atmospheric warming could result in the release of C as CO_2–C, previously held within the soil organic matter stock, back to the atmosphere. At the same time, there is a lack of agreement about how soil methane fluxes would respond to changes in soil conditions. For example, variable soil methane emissions have been reported with respect to different temperatures (Christensen et al., 1997; Rustad and Fernandez, 1998; Del Grosso et al., 2000). Carbon rich soils such as those found in grassland, peatland and forest ecosystems that are currently reservoirs of terrestrial C and biodiversity and could thus have considerable potential as sources of greenhouse gases under warmer conditions.

It has been estimated that soil heterotrophic respiration and CO_2 production doubles with every 10 °C increase in atmospheric temperature, i.e. $Q_{10}=2$ (Sarmiento, 2000) with serious implications for the carbon-climate phenomenon. In contrast, Luo et al. (2001) found that soil respiration showed only a short lived response to warmer temperature regimes suggesting that heterotrophic 'acclimatization' would eventually reduce the feed-back effect of soil C release on climatic change. The primary causes of this observed reduction in soil respiration under warmer climates are decreased soil moisture content resulting in diminished root and microbial respiration (Peterjohn et al., 1994; Atkin et al., 2000) and progressive C substrate limitation (Rustad and Fernandez, 1998; McHale et al., 1998). Therefore the sensitivity and resilience of terrestrial ecosystems to shifts in environmental conditions anticipated as a result of climate change will rely upon the responses of soil organisms to temperature and moisture changes.

Enchytraeids are often the most abundant faunal component in upland grassland soils in the UK and can comprise up to ca. 70% of the total animal biomass in some ecosystems (Cragg, 1961). Populations are frequently dominated by *Cognettia sphagnetorum* constituting up to 95% of the total enchytraeid biomass (Coulson and Whittaker, 1978). Climate change studies have demonstrated the sensitivity of this species to temperature and moisture regimes in terms of reproduction rates and vertical migration (Briones et al., 1997), with important implications for nutrient cycling (Briones et al., 1998a,b; Cole et al., 2002a). The contribution of enchytraeid respiration to soil CO_2 emissions has been reported as being between 2 and 40% of total soil respiration (Satchell, 1971; Didden, 1993). However, indirect effects based on the interaction of enchytraeids with other soil organisms (e.g. bacteria and fungi) could be more important when determining ecosystem responses to global change. It is already known that feeding activities of enchytraeids increase soil microbial activity (total and fungal) by 35% in blanket peat (Cole et al., 2000) and that warming promotes enchytraeid grazing on soil microorganisms leading to decreases in microbial biomass which in turn reduces their ability to immobilise soil nutrients (Cole et al., 2002b). Determining the direct and indirect effects of increased temperature on soil biology and subsequent C release remains a priority.

In this study we examined the link between enchytraeid activity and biomass on soil C fluxes across a range of temperatures (that reflected the natural ranges observed in the field over the course of a year). The work had two components: (i) an investigation into the effects of elevated temperatures and enchytraeid activity on C fluxes and (ii) to develop an empirical model to predict soil CO_2 production in two different organic upland grassland soils at different temperatures.

2. Material and methods

2.1. Soil and experimental set-up

Soil cores (10 cm deep×5 cm diameter) were taken in May 2001 from grassland plots located on Rigg Foot Hill, Sourhope Farm, Scotland (2° 5′ W 55° 30′ N). The site is an unimproved upland grassland that was grazed by sheep until April 1998, when stock was excluded. The vegetation is dominated by an *Agrostis capillaris* (L.), *Festuca ovina* (L.) community typical of upland grassland across UK. The soil is classified as originating from a brown forest earth (cambisol) sandy silt loam formed on glacial till derived from andesitic lava. It has a high organic matter content with 7.6±0.8% C and 0.56±0.04% N and a mean soil pH that is low 4.7±0.2 (Ostle et al., 2000).

Intact soil cores were brought to the laboratory where surface vegetation was removed and the soil profile sliced into 5 cm horizontal layers. The uppermost 0–5 cm deep rhizosphere layer (O/H horizon) was used in the current experiment. Soil was sieved through a 2 mm mesh and living plant material (root, stem, leaf and stolon) was removed by hand. Mesofauna extractions made on sieved soil indicated that this procedure had effectively eliminated the native enchytraeid populations. Moisture content was adjusted to give 60% water as calculated on a dry weight basis. A series of 24 Wheaton bottles (100 cm^3) were used as C-release microcosms with each receiving 10 g of sieved fresh soil. Half of the microcosms were inoculated with enchytraeids (see below) and designated as +enchytraeid treatments (+E) with the remaining twelve used as control treatments (C). Four replicate microcosms were incubated, in darkness, at each of the three temperature treatments 4, 10 and 15 °C for a total of 116 days.

Enchytraeids were extracted from intact 0–5 cm slices of soil from the cores described above using a modified wet funnel method (O'Connor, 1955). Next, 25 individuals were inoculated into each +E microcosm, i.e. to give

a population density of 2.5 enchytraeid individuals per gram of soil (fresh weight) in four replicates of each temperature treatment (4, 10 and 15 °C). This number of enchytraeids was comparable to that observed at the Sourhope grassland field site at the time of sampling. Soil moisture content was continuously monitored and maintained at 60% (dry weight basis) in all microcosms, by means of regular weighing and addition of purified water.

2.2. Enchytraeid biomass and numbers

Changes in enchytraeid populations during incubation were determined using a parallel series of microcosm units maintained at 4, 10 and 15 °C until required for destructive sampling for mesofauna extraction. Plastic containers (50 cm^3) were filled with 20 g of the fresh sieved soil, resulting in a total of 120 experimental units (4 replicates × 2 fauna treatments × 3 temperature treatments × 5 sampling dates). On day 3, half of them were inoculated with 50 enchytraeid specimens extracted as described above (to give a population density of 2.5 individuals g $soil^{-1}$) with the remaining 60 referred as controls. Soil was maintained at 60% moisture content (dry weight basis). On each sampling occasion, 16, 33, 44, 65 and 86 days after the start of incubation, four replicates of each temperature and faunal treatments were extracted separately and total numbers and dry weights (freeze-dried −20 °C) of enchytraeids collected were determined.

2.3. CO_2 and CH_4 flux measurements

CO_2 and CH_4 fluxes were measured from mesofauna (+E) and control (C) temperature treatments at 3, 16, 33, 44, 65, 86, 116 days. On each sampling occasion Wheaton bottle microcosms were sealed and 5 ml of headspace gas was removed after 10 min to be replaced with 5 ml of an air standard of known CO_2 and CH_4 concentrations. This was done to ensure that microcosms were maintained at atmospheric pressure throughout their incubation period. A second 5 ml sample of headspace gas was then taken after 180 min to enable determination of flux rates. Gas samples were analysed for CO_2 and CH_4 concentrations using a Gas Chromatograph fitted with an FID and methaniser (AutoSystem XL, Perkin Elmer) immediately after sampling. After the subtraction the 5 ml standard volume replacement gas fluxes were calculated as μg CO_2–C and μg CH_4–C g $soil^{-1}$ day^{-1} using the difference between the 10 and 180 min interval concentrations, relative to a reference gas standard. Flux results were also expressed in relation to enchytraeid dry weight biomass i.e. CO_2–C or CH_4–C μg mg enchytraeid $tissue^{-1}$ day^{-1}.

2.4. Statistical analyses

One-way ANOVA was applied to independent measures of enchytraeid numbers and biomass to test for significant differences between temperature treatments. Separation of means was determined using Tukey's Studentized range (HSD) test ($\alpha = 0.05$).

Gas emissions (CO_2–C and CH_4–C production were related to dry weight of soil and of mesofauna biomass) from different temperature treatments over time and were analysed by repeated-measures ANOVA. Mauchly's test (Crowder and Hand, 1990) was carried out and determined that the data did not meet the assumption of a common spherical covariance matrix and, for that reason, the results were interpreted by using the multivariate output of repeated measures ANOVA. In order to study the pattern of the time response the analysis of repeated-measures was performed by including the 'profile' option in the analysis in which differences are constructed from adjacent levels of the Time factor. In addition, in order to avoid error due to missing values for CH_4 fluxes, data from the two first sampling occasions were not included in the statistical analyses.

Regression analyses of individuals-biomass, temperature-biomass and biomass-CO_2 production are presented as R, p, n. The functions obtained were then used to produce a two step regression model to predict soil CO_2 production in two field sites, Sourhope grassland soil and Great Dun Fell moorland soil, with known enchytraeid biomass. In this model predicted biomass and predicted total respiration were calculated at +2.6 and +5.0 °C (above ambient temperature) at both sites. The validity of using temperature to predict changes in enchytraeid biomass was assessed using atmospheric temperature and enchytraeid data obtained from a warming experiment made in the field at Great Dun Fell (Briones et al., 1997, 1998b).

All statistical analyses were performed on the data using SAS system. Differences at the $P < 0.05$ level were considered significant.

3. Results

3.1. Community structure

The initial enchytraeid inoculum obtained from the Sourhope grassland soil was dominated both in numbers and biomass by *Cognettia sphagnetorum* (Vejdovsky) (89%), with some *Achaeta* sp. Vejdovsky (9%) and a few specimens of *Fridericia* sp. Michaelsen (2%) also present. Community composition remained the same in all temperature treatments for the duration of the 116 day experimental incubation period.

3.2. Numbers and biomass

Defaunation resulted to be a successful method and enchytraeid extractions from the control units at each sampling occasion showed that only few worms survived

the sieving process and this low density remained so for the entire experimental period.

In the units with enchytraeids numbers and biomass increased during the course of the experiment at all three temperature treatments; 4, 10 and 15 °C (Fig. 1). The mean initial rate of increase (i.e. up to 33 days) in enchytraeid numbers and biomass was 0.16 ± 0.04 individuals g soil^{-1} day^{-1} and 6.1 ± 0.3 μg g soil^{-1} day^{-1}, respectively. There was no significant difference in the rate of increase between treatments. At the highest temperature (15 °C) this increase was significantly greater than that seen at the two lower temperature treatments after 65 days (Numbers: F-value = 13.04, $P < 0.01$; Biomass: F-value = 5.76, $P < 0.05$). By day 86 enchytraeid numbers and biomass in the 15 °C treatment had increased to over threefold initial values (day 3) (Numbers: F-value = 27.40, $P < 0.0001$; Biomass: F-value = 22.36, $P < 0.0003$). However, enchytraeid numbers and biomass in the 4 and 10 °C treatments were seen to stabilise at 8.5 ± 0.3 individuals g soil^{-1} and 0.4 ± 0.01 mg g soil^{-1} after 33 days and there was no significant difference between 4 and 10 °C treatments over the course of the experiment. The relationship between enchytraeid biomass and numbers of individuals per gram of soil was statistically significant ($R = 0.76$, $P < 0.01$, $n = 84$).

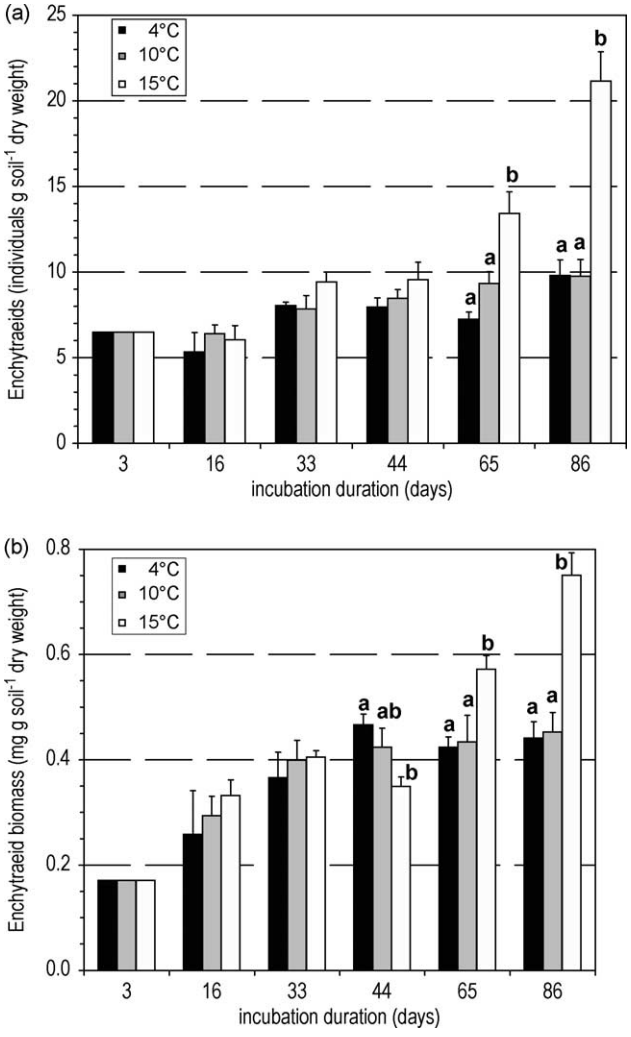

Fig. 1. Changes in enchytraeid populations during the course of the 86 days incubation period: (a) numbers (individuals g soil^{-1}) and (b) biomass (mg g soil^{-1}) dry weight basis. Means and standard error values are shown for 4, 10 and 15 °C temperature treatments.

3.3. CO_2–C fluxes

Table 1 shows the results of the multivariate approach of repeated measures ANOVA for the overall mean CO_2–C production (μg g soil^{-1} day^{-1}). Temperature, enchytraeid presence and their combined influence had a significant effect on CO_2–C production. In addition CO_2–C fluxes significantly changed with time and this change across different sampling dates depended on temperature (Table 1).

In Fig. 2a CO_2–C production (μg g soil^{-1} day^{-1}) in control (C) treatments at all incubation temperatures is shown. The results of repeated-measures ANOVA showed a significant temperature effect over time ($F = 14.48$, $P = 0.0083$). Measurements of gas fluxes made after 16 days showed an initial priming effect in the 10 and 15 °C treatments ($P < 0.05$) followed by stabilisation between 20 and 40 μg g soil^{-1} day^{-1} by day 65 (with highest fluxes produced at 15 °C). At the final 116 day sampling differences in CO_2–C fluxes between control temperature treatments were not significant.

In the enchytraeid treatments (+E) CO_2–C production (μg g soil^{-1} day^{-1}) also showed a priming effect ($P < 0.05$) in the 10 and 15 °C treatments at day 16 (Fig. 2b). By day 44 CO_2–C emissions had stabilised between 20 and 40 μg g soil^{-1} day^{-1} in the 4 and 10 °C treatments whereas CO_2–C fluxes maintained at 15 °C ranged between 40 and 55 μg g soil^{-1} day^{-1}. The results of repeated-measures ANOVA showed a significant temperature effect ($F = 79.35$, $P = 0.0001$) with CO_2 production in the 15 °C treatment being 1.5 times greater than that recorded at the 4 °C treatment on day 116 ($P < 0.05$).

When comparing animal treatments at each temperature level, the average values of CO_2–C production (μg g soil^{-1} day^{-1}) at 15 °C were significantly

Table 1
Results from MANOVA (between- and within-subject factors) for CO_2–C production (μg g soil^{-1} day^{-1})

Source of variation	F	df	P
Between-subjects			
Temperature	74.77	2	0.0001
Enchytraeids	22.26	1	0.0004
Temperature × enchytraeids	9.74	2	0.0026
Within-subjects			
Time	18.6252	6	0.0003
Time × temperature	22.6955	12	0.0001
Time × enchytraeids	0.6819	6	0.6704
Time × temperature × enchytraeids	1.4949	12	0.2231

Significance multivariate test on each is Wilks' lambda test.

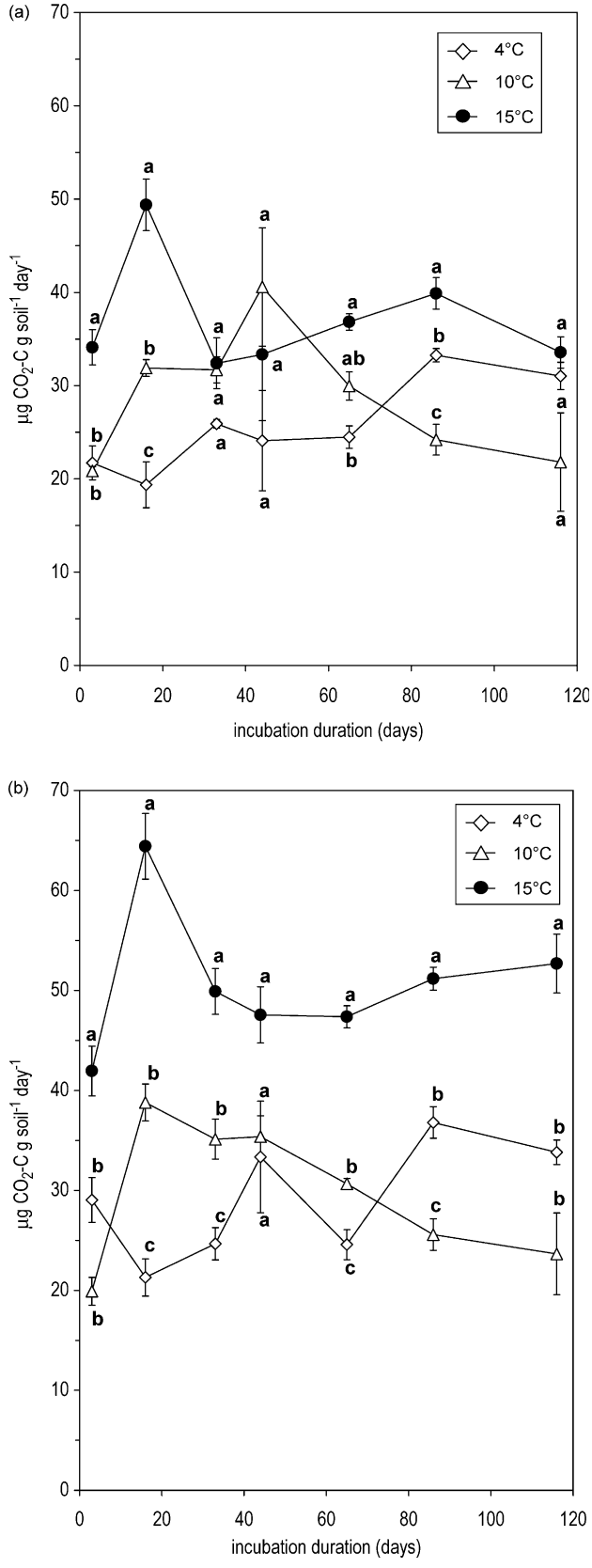

Fig. 2. CO_2–C production (µg g soil^{-1} day^{-1}) from (a) control soil (without enchytraeids) and (b) total soil (with enchytraeids) during the course of the 116 days incubation experiment. Means and standard error values are shown for 4, 10 and 15 °C temperature treatments.

higher in the enchytraeid treatments than those in the controls ($F=30.80$, $P=0.0026$) whereas no significant differences were detected at the two lower temperatures. In addition, total soil respiration in the +E 15 °C treatment was significantly higher than in the controls for the entire incubation period ($P<0.05$), with the exception of two sampling occasions (3 and 44 days).

The contribution of enchytraeids to CO_2–C production is the result of their direct (metabolism) and indirect effects (changing the physical and chemical characteristics of the soil environment and interacting with microbial populations). This can be calculated as the difference between total soil CO_2–C and control soil CO_2–C production (Fig. 3). After day 44 enchytraeid activities significantly increased the amount of CO_2–C produced (µg g soil^{-1} day^{-1}) at the highest temperature tested, whereas no significant differences were detected at 4 and 10 °C. The results from the multivariate Wilk's test showed that time ($F=60.60$, $P=0.0032$), temperature ($F=22.92$, $P=0.0005$) and the combined influence of these two factors ($F=21.59$, $P=0.0006$) had a significant effect on CO_2 production associated to enchytraeid activity.

In order to determine to what extent this enchytraeid induced respiration accounted for the total CO_2–C produced, the percentage of their contribution to the total production was also calculated (Table 2). The results of

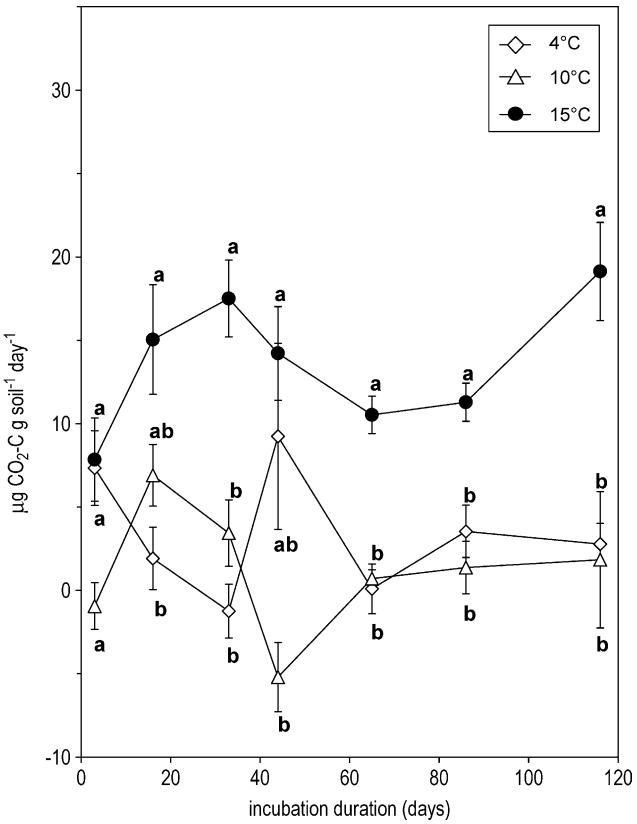

Fig. 3. CO_2–C production (µg g soil^{-1} day^{-1}) associated to enchytraeid activity (calculated as the difference between total soil CO_2–C and control soil CO_2–C production).

Table 2
Results from the repeated measures ANOVA for the percentage contribution of enchytraeid induced CO_2–C production (i.e. direct + indirect) to total soil CO_2–C production during the 116 days incubation period

Temperature	Incubation (days)							Means
	3	16	33	44	65	86	116	
4 °C	25.3±7.7a	9.0±8.8a	−5.0±6.6a	27.7±16.7a	0.4±6.1a	9.6±4.3a	8.2±3.7a	10.74±4.5a
10 °C	−4.7±7.0a	17.8±4.8a	9.8±5.7a	−14.7±5.9a	2.3±1.7a	5.4±6.2a	7.8±17.3a	3.37±4.0a
15 °C	18.7±5.9a	23.3±5.1a	35.1±4.6b	29.9±5.9a	22.2±2.4b	22.1±2.2a	36.3±5.6a	26.80±2.6b

Calculation of percentage contribution of enchytraeid induced respiration (%E) to total respiration; $\%E = E \div (T \div 100)$, where E, enchytraeid induced CO_2–C (μg g soil^{-1} day^{-1}) and T, total soil respiration CO_2–C (μg g soil^{-1} day^{-1}) from enchytraeid treatments. Values are means with standard errors ($n=4$) of three temperature treatments, 4, 10 and 15 °C. Means with the same letters are not significantly different (Tukey grouping, $P<0.05$).

the multivariate approach of the repeated measures analysis showed that there was no time effect but temperature significantly influenced the percentage of total respiration associated with enchytraeids. This resulted in an increase in CO_2 fluxes of 10.74±4.5, 3.37±4.0 and 26.80±2.6% in 4, 10 and 15 °C treatments compared to controls (Table 2).

3.4. CH_4–C fluxes

Measurements of CH_4 fluxes from the incubation treatments showed that the grassland soil was a strong consumer of CH_4 and that there was little change in CH_4 consumption in all treatments during the course of the 116 day experimental period. Neither temperature, enchytraeids, nor the interaction between these two factors had any statistically significant effect on overall mean CH_4–C fluxes, however, time and the interaction between time and temperature had a significant effect on methane fluxes (Table 3). The profile analysis applied to each of the contrast variables (Table 4) showed that a significant change in CH_4 fluxes occurred between 44 and 86 days.

3.5. Gas fluxes and enchytraeid biomass

Total soil CO_2 production (+E) was also found to be positively linked with enchytraeid biomass ($R=0.76$, $P<0.05$, $n=84$) with a mean production at the end of the experimental period of 77.01±6.05 CO_2–C μg mg enchytraeid tissue^{-1} day^{-1} ($n=12$) irrespective of the temperature treatment. The results of the multivariate approach of repeated measures ANOVA for CO_2–C and CH_4–C productions per unit of enchytraeid biomass (CO_2 or CH_4 C μg mg enchytraeid tissue^{-1} day^{-1}), over six and four sampling dates, respectively, are shown in Table 5. Time had a significant effect on greenhouse gas emissions but the interaction between time and temperature was only significant for CO_2–C production. No temperature effect was found for methane fluxes whereas a positive influence of temperature on net CO_2–C flux per unit enchytraeid biomass was observed. And thus, until day 65 and with the exception of two sampling dates (16 and 33 days), CO_2 production/unit biomass was significantly higher

Table 3
Results from MANOVA (between- and within-subject factors) for CH_4–C production (μg g soil^{-1} day^{-1})

Source of variation	F	df	P
Between-subjects			
Temperature	1.31	2	0.2995
Enchytraeids	0.35	1	0.5649
Temperature × enchytraeids	1.90	2	0.1840
Within-subjects			
Time	84.1140	4	0.0001
Time × temperature	2.6260	8	0.0320
Time × enchytraeids	0.6896	4	0.6130
Time × temperature × enchytraeids	1.2823	8	0.2985

Significance multivariate test on each is Wilks' lambda test.

Table 4
ANOVAs on each of the contrast variables of the time factor for CH_4–C production (μg g soil^{-1} day^{-1})

Source of variation	df	MS	F	P
Contrast variable week 7 to week 5				
Mean	1	0.0046	3.98	0.0647
Temperature	2	0.0010	0.91	0.4242
Enchytraeids	1	0.0017	1.50	0.2389
Temperature × enchytraeids	2	0.0002	0.18	0.8389
Error	15	0.0011		
Contrast variable week 10 to week 7				
Mean	1	0.0251	12.63	0.0029
Temperature	2	0.0024	1.22	0.3222
Enchytraeids	1	0.0005	0.28	0.6058
Temperature × enchytraeids	2	0.0000	0.01	0.9919
Error	15	0.0020		
Contrast variable week 13 to week 10				
Mean	1	0.0263	29.62	0.0001
Temperature	2	0.0022	2.51	0.1149
Enchytraeids	1	0.0000	0.06	0.8128
Temperature × enchytraeids	2	0.0002	0.24	0.7881
Error	15	0.0009		
Contrast variable week 17 to week 13				
Mean	1	0.2120	1.20	0.2906
Temperature	2	0.2545	1.44	0.2678
Enchytraeids	1	0.0570	0.32	0.5785
Temperature × enchytraeids	2	0.2392	1.35	0.2880
Error	15	0.1767		

Table 5
Results from MANOVA (between- and within-subject factors) for CO_2–C and CH_4–C productions per unit of enchytraeid biomass (μg mg enchytraeid tissue^{-1} day^{-1})

Source of variation	*F*	d*f*	*P*
CO_2–C production			
Between-subjects			
Temperature	38.08	2	0.0001
Within-subjects			
Time	34.9281	5	0.0007
Time×temperature	6.5920	10	0.0031
CH_4–C production			
Between-subjects			
Temperature	2.64	2	0.1254
Within-subjects			
Time	49.1577	3	0.0001
Time×temperature	2.2547	6	0.0986

Significance multivariate test on each is Wilks' lambda test.

in the 15 °C treatment than in the other two temperature regimes (Fig. 4). Following this CO_2–C flux per unit enchytraeid biomass decreased and reached similar values to the ones measured at the lower temperature regimes.

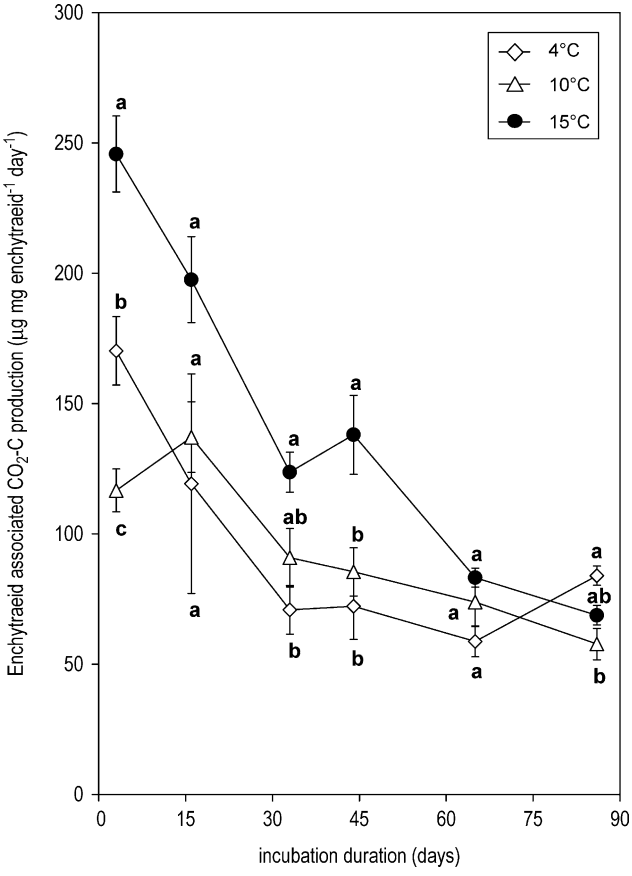

Fig. 4. CO_2–C production associated to enchytraeid biomass (calculated as the difference between total soil CO_2–C and control soil CO_2–C production) per unit of enchytraeid dry weight biomass (μg mg enchytraeid tissue^{-1} day^{-1}).

3.6. Potential enchytraeid induced soil CO_2 production in the field

The positive relationship between temperature, enchytraeid biomass and total soil respiration (Fig. 4) observed in the microcosm experiment (i.e. 77.01 ± 6.05 CO_2–C μg mg enchytraeid tissue^{-1} day^{-1}) was used to produce a two step regression model to predict soil CO_2 production (CO_2–C mg^{-1} m^2). Enchytraeid biomass = (0.05 × temperature) − 0.0591; $R = 0.78$, $P < 0.04$, $n = 12$, and soil respiration = (62.802 × biomass) + 3.6508; $R = 0.65$, $P < 0.05$, $n = 84$. The model was applied to two different organic upland grassland soils with known enchytraeid biomass; Sourhope grassland soil (7–10% C) and Great Dun Fell moorland soil (30% C). The validity of using temperature to predict changes in enchytraeid biomass (tissue dry weight g^{-1} m^2) was assessed using atmospheric temperature and enchytraeid data obtained from a warming experiment made in the field at Great Dun Fell (Briones et al., 1997). Results showed that a 5 °C increase in atmospheric temperature above mean ambient temperature could have the potential to produce a significant increase in enchytraeid biomass resulting in a near twofold increase in soil CO_2 release from both soil types.

4. Discussion

Since enchytraeids are burrowing animals and ingest large amounts of soil their activities have an important influence on other components of the soil community. In this study the respiration values of all soil heterotrophs in the presence of and without enchytraeids over time showed that more CO_2 was released from the soils when these organisms were present and this effect was most pronounced at the highest temperature (15 °C). Therefore we conclude that the presence of an active enchytraeid community and its interaction with temperature exerted a significant influence on CO_2 emissions.

In contrast to CO_2 production, no significant relationships were observed between net CH_4 fluxes and temperature. This could be explained by the fact that moisture was maintained constant throughout the experimental period; however, temperature is expected to have important effects on the water table in the field, which in turn will influence CH_4 oxidation.

Among ecosystem responses reported in the literature, soil respiration and CO_2 production, show a consistent response to temperature, with warming leading to an increase in CO_2 released to the atmosphere. For example, Updegraff et al. (2001) found that in mesocosm studies seasonal ecosystem respiration responded almost exclusively to temperature and did not differ between community types or with water table levels. However, when extrapolating this result to global scales differences in spatial (greater response in warmer ecosystems)

and temporal (transient response) patterns are observed (Rustad et al., 2001). Clearly, in the field, interactions between precipitation and soil moisture will play a critical role in the coupling between atmospheric temperature and soil respiration.

Previous research (Luo et al., 2001) showed no significant increase in respiration from a tall grass prairie soil maintained at 2–2.6 °C above ambient temperatures, suggesting that heterotrophic 'acclimatization' could be an important factor in reducing soil CO_2 release at lower temperatures, in particular in those ecosystems with low soil carbon storage. In contrast, Sarmiento (2000) predicted a 100% increase in soil CO_2 production with every 10 °C (i.e. $Q_{10}=2$), which is comparable with our results at lower temperatures (4–10 °C), both in the presence and absence of enchytraeids (i.e. $Q_{10}+E=1.7$, $Q_{10}-E=1.9$). Furthermore, in our experiment the greatest increase in CO_2 production was observed at temperatures above 10 °C with Q_{10} highest in the presence of enchytraeids ($Q_{10}+E=3.4$), representing a 25% increase in soil respiration when compared to the defaunated systems ($Q_{10}-E=2.6$). This confirms the findings of McHale et al. (1998) who suggested that total soil respiration could be even stronger at higher temperatures. Therefore, taken together these results suggest that in organic soils such as grasslands and peatlands, the impacts of soil warming on frequently large enchytraeid populations and their interactions with microbial activities can be important.

The impacts of atmospheric warming on ecosystem C function are mediated directly by faunal and microbial communities through their feeding activities or indirectly through their role in changing the physical and chemical characteristics of the soil environment. Enchytraeids are known to ingest mineral particles, fungi, bacteria, oats and yeasts, algae, and even dead bodies of lumbricids and arthropods, however other work suggest that they are 80% microbivorous and 20% saprovorous (Didden, 1993). It is widely accepted that soil fauna-microorganisms interactions result in enhanced mineralisation and nutrient release (Bardgett et al., 1998; Bardgett and Chan, 1999; Cole et al., 2000) however, these effects could be altered under warmer scenarios either by changes in soil biota biomass and/or their activities (Cole et al., 2002b).

Briones et al. (1997) found that individual species respond differently to changes in temperature by exhibiting different strategies. And for instance, *C. sphagnetorum* (our dominant species here) was sensitive to temperature and moisture regimes in the field resulting in vertical migration away from dry top soil and reproduction by fragmentation being stimulated by increased temperatures. If temperatures rise and precipitation increases during the summer months across Northern Europe, as foreseen by some studies (e.g. Sala et al., 2000), then we will see an increase in enchytraeid numbers and biomass in organic soils (with soil moisture content and temperature maintained within a range suitable for the survival and reproduction of enchytraeids). Such increases in numbers were seen in the Great Dun Fell-Sink Beck soil transplanting experiment (Briones et al., 1997) that produced a 2.6 °C warming of the soil. Similarly, Cole et al. (2002a) predicted a 43% increase in enchytraeid abundance for a 2.5 °C increase in mean monthly temperature at Moor House. Clearly such temperature induced changes in enchytraeid numbers and biomass would have considerable potential to increase soil CO_2 production (as observed in the current experiment) that could, in turn, contribute further to climate forcing. Indeed the results of our simple regression model linking atmospheric temperature, enchytraeid biomass and total soil CO_2 release suggest that if soil moisture levels remain compatible with enchytraeid growth and reproduction then soil warming could produce important increases in soil CO_2 release. And thus a 5 °C increase in atmospheric temperature could result in a near twofold increase in soil CO_2 release. This model assumes that the role of enchytraeids in this process is biomass dependent and that biomass increases will be more important than changes in functional group dominance within the soil mesofauna community. It is also important to consider that the potential for enchytraeid biomass to increase and remain elevated in the field will be regulated by temperature induced changes on plant growth and carbon inputs and by nutrient constraints. It is therefore likely that there will be some acclimatization or stabilisation of the enchytraeid biomass as other conditions and resources become limiting. Clearly the useful application of this form of model in the future will be reliant on the inclusion of other growth-rate limiting conditions (e.g. hydrology, vegetation effects, changes in nutrient availability).

In conclusion, our results indicate that enchytraeids exerted a positive influence on C release from organic soils and therefore they offer a representative and abundant group that through their activities can have important impacts upon total soil heterotroph respiration. The methodology adopted in the incubation experiment described here could be considered too simplistic for extrapolation to the field scale as the effects of other important components of the soil system (e.g. plants and a more complex food web) have not been considered in the experiment. Nonetheless, the results obtained here do provide evidence of the sensitivity of certain soil mesofauna groups to ecological perturbation and environmental change such as atmospheric warming and demonstrate the important influence of enchytraeids upon CO_2 production in response to temperature. Therefore, we suggest that enchytraeids do have potential as measurable indices of biological sensitivity to climatic changes that could be used to monitor and predict the fate of soil C in the face of environmental change. This is particularly true of moorland grassland and peatland soils in the Northern hemisphere that have been accumulating atmospheric CO_2 via photosynthesis since the last ice age ($\sim$10,000 years).

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