



Residue incorporation and N fertilization affect the response of soil nematodes to the elevated CO₂ in a Chinese wheat field

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ABSTRACT

The interplay between the carbon and other nutrient cycles is the key to understand the responses of soil ecosystems to climatic change. Using the free-air CO₂ enrichment (FACE) techniques, we carried out a multifactorial experiment in a Chinese rice–wheat rotation system, to investigate the response of soil nematodes to elevated CO₂ under different application rates of N fertilizer (225.0 kg N ha⁻¹ (HN) and 112.5 kg N ha⁻¹ (LN), respectively) and residue incorporation (0 kg C ha⁻¹ (ZR), 1000 kg C ha⁻¹ (MR) and 2000 kg C ha⁻¹ (HR), respectively). This study was conducted during the wheat growing season of 2007 after exposure to the elevated CO₂ for three years. The results in our study indicated that seasonality is an important factor in determining changes in the nematode abundance and diversity. The residue addition effects were more obvious than the elevated CO₂, which significantly influenced the abundance of total nematodes and plant-parasites, and some ecological indices. The interactions between residue addition and CO₂ significantly influenced nematode dominance and structure indices. High level of N fertilization was found to decrease the nematode diversity, generic richness and maturity indices at wheat jointing stage. There are significant interactions between N fertilization and elevated CO₂ for abundance of total nematodes and different trophic groups.

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1. Introduction

Atmospheric CO₂ concentrations have increased since industrialization and are projected to increase further (IPCC, 2007), which will greatly affect physiology of different plants and organisms. In addition to the direct impacts on aboveground vegetation, the climate change may also influence belowground processes and affect nutrient turnover in soils (Rogers et al., 1994; Sowerby et al., 2005). Since soil decomposer animals exert a strong regulatory control on soil structure and soil processes, such as decomposition activity and nutrient cycling (Haimi et al., 2005), it is important to assess the responses of soil fauna to the elevated CO₂ in order to fully understand potential effects of climate change (Hoeksema et al., 2000; Yeates et al., 2003; Haimi et al., 2005).

The interplay between the carbon and other nutrient cycles is the key to understand the responses of terrestrial ecosystems to climatic change. Increased C and N availability has been reported to increase plant biomass (Daepf et al., 2000), modify plant C

partitioning (Cotrufo and Gorissen, 1997), and alter plant tissue C/N ratio which tends to alter the decomposability of plant residues (Van Groenigen et al., 2005). However, all these potential responses may be constrained by belowground processes and mediated by responses of soil biota (Jones et al., 1998; Coûteaux and Bolger, 2000; Frederiksen et al., 2001; Ge et al., 2008).

Since soil nematodes form one of dominant belowground communities in agroecosystems (Ritz and Trudgill, 1999), the study of their responses can help us better understand agroecosystem response to climate change. Researches incorporating the soil fauna indicated that elevated atmospheric CO₂ can significantly alter the structure of the soil food web (Yeates et al., 1997, 2003; Hungate et al., 2000; Niklaus et al., 2003; Neher et al., 2004; Sonnemann and Wolters, 2005; Haimi et al., 2005). However, Ayres et al. (2008) recently found that belowground nematode herbivores were resistant to elevated CO₂ in grassland ecosystems. While there are a variety of reports on the response of soil biota to elevated CO₂, relatively few studies have examined the impacts of residue incorporation and N fertilization on the responses of soil biota to the elevated CO₂ (Moscatelli et al., 2005; Sticht et al., 2006; Lagomarsino et al., 2007; Garcia et al., 2008). Some researchers

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have reported that the addition of plant residue or the application of inorganic amendments could lead to the changes in nematode communities, such as an increase or decrease in abundance and the alterations of community structure (Ferris and Matute, 2003; Wang et al., 2004; Chen et al., 2007; Okada and Harada, 2007; Liang et al., 2009). As the interactions between carbon (C) and nitrogen (N) are likely to modulate terrestrial ecosystem responses to elevated CO₂ at different scales (Reich et al., 2006), we hypothesized that changes in the supplies of nitrogen fertilization and decomposable organic carbon would modify the effects of enriched atmospheric CO₂ on soil nematodes.

Using the FACE experimental setup, we evaluated the response of soil nematodes to the elevated CO₂ in a Chinese wheat field with different application rates of N fertilization and residue incorporation. Our specific objectives were to answer following two questions: (1) if the residue incorporation or different levels of N fertilization influences the response of soil nematodes to the elevated CO₂; and (2) how their interactions affect soil nematode responses.

2. Materials and methods

2.1. Experimental site and FACE facilities

The experimental site is located in a suburb of Jiangdu city in Jiangsu province of China (32°35'N, 119°42'E). The soil at the study site is a Shajiang-Aquic Cambosol, with 18.4 g kg⁻¹ total C, 1.5 g kg⁻¹ total N, pH (H₂O) 7.9, 13.7% clay (<0.002 mm) and bulk density 1.16 g cm⁻² at 0–15 cm depth (Li et al., 2007). An experimental platform of free-air CO₂ enrichment (FACE) was established in mid-June of 2004 over a rice–wheat rotation system, with rice transplanted in mid-June and harvested in middle-to-late October and winter wheat was sown in early November and harvested in late May or early June of the next year. Three replicate FACE rings, each with a size of 80 m², were set randomly within a uniform area of 200 m × 400 m to continuously provide an elevated CO₂ concentration of 200 ± 40 μmol CO₂ mol⁻¹ over the ambient conditions (this setup is hereinafter referred to as FACE), while three replicate rings, each with the same size, were set randomly within the same area for the ambient CO₂ treatment (hereinafter referred to as Ambient). All of the rings were far enough apart to prevent CO₂ from spilling over from one ring to another and to avoid the influence of CO₂ from the FACE rings on the Ambient plots, the details of the FACE facilities are described by Liu et al. (2002).

2.2. Nitrogen and decomposable organic carbon additions

In each FACE and Ambient plot, two levels of N fertilization were applied in each sub-plot. Ammonium-based nitrogen fertilizer was applied in the wheat season at the rates of high nitrogen treatment (HN) with 350.0 kg N ha⁻¹ and low nitrogen treatment (LN) with 174.0 kg N ha⁻¹, respectively. For the HN and LN treatments, 50% was applied basally, and 10% and 40% was top-dressed at early- and late-tillering, respectively. Each replicate treatment of nitrogen addition was assigned on a plot of 20–30 m². Ammonium-based nitrogen fertilizers were used in both wheat and rice cropping seasons. Phosphorus and potassium fertilizers were applied in all plots at rates of 75 kg P₂O₅ ha⁻¹ and 75 kg K₂O ha⁻¹ in either rice or wheat season. Each sub-plot receiving high and low nitrogen treatment was subdivided into sub-sub plots receiving residue at three different addition rates under the elevated and ambient CO₂, i.e. 0 kg C ha⁻¹ (hereinafter referred to as ZR), moderate addition rate of 1000 kg C ha⁻¹ (hereinafter referred to as MR) and high addition rate of 2000 kg C ha⁻¹ (hereinafter referred to as HR), respectively. Before the first rice season in 2004, one sub-plot of 1 m² was set for ZR, and another of the same area was set for HR, while the remaining area of

each HN and LN plots was treated with MR. For the MR and HR treatments, rice straw from the previous rice season was incorporated in which the rice was growing. The harvested rice straws for the ZR treatment were completely removed from the experimental fields. No additional organic matter was incorporated in the wheat growing seasons. Different treatments were established in 2004 and have been maintained since then. This experiment was conducted during the wheat growing season of 2007, after expose to elevated CO₂ for three years.

2.3. Sampling, extraction and identification of nematodes

Soil samples were collected from 0 to 15 cm depth in each plot, at jointing and ripening stages of wheat growing seasons (March 25 and May 28 in 2007, respectively). Each soil sample pooled from five soil cores (2.5-cm diameter) was stored in individual plastic bags, and transferred to a 4 °C cold room.

Nematodes were extracted from 100 g soil sample (fresh weight) by a modified cotton–wool filter method (Oostenbrink, 1960; Townshend, 1963; Liang et al., 2009). Nematode populations are expressed as number of nematodes per 100 g dry soil and at least 150 nematodes from each sample were identified to genus level using an inverted compound microscope. The trophic groups of soil nematodes characterized by feeding habits and life-history characters were assigned to: (1) bacterivores (Ba); (2) fungivores (Fu); (3) omnivores–carnivores (Om + Ca) and (4) plant-parasites (Pl) (Yeates et al., 1993).

2.4. Nematode community analyses

Nematode ecological indices were calculated by the following approaches: (1) trophic diversity $TD = 1/\sum p_i^2$; where p_i is the proportion of trophic group i ; (2) Shannon–Weaver diversity $H' = -\sum p_i(\ln p_i)$; (3) dominance index $\lambda = \sum p_i^2$; (4) generic richness $SR = (S - 1)/\ln(N)$, where p_i is the proportion of individuals in the i th taxon, S is the number of taxa and N is the number of total nematodes (Yeates and Bongers, 1999); (4) maturity index $MI = \sum v(i) \cdot f(i)$, where $v(i)$ is the c - p value of taxon i according to their r and K characteristics following Bongers (1990), $f(i)$ is the frequency of taxon i in a sample; (5) plant parasite index (PPI) which was determined in a similar manner for plant-parasitic genera (Bongers, 1990); (6) structural index $SI = 100 \times (\sum k_s n_s / (\sum k_s n_s + \sum k_b n_b))$, (7) enrichment index $EI = 100 \times (\sum k_e n_e / (\sum k_e n_e + \sum k_b n_b))$, where k_b is the weight assigned to guilds Ba₂ and Fu₂ and n_b is the abundance of nematodes in guilds Ba₂ and Fu₂, which indicate basal characteristics of the food web; k_s the weight assigned to guilds Ba₃–Ba₅, Fu₃–Fu₅, Om₄–Om₅ and Ca₂–Ca₅, n_s is the abundance of nematodes in these guilds, which represent the structure condition of the food web; k_e the weight assigned to guilds Ba₁ and Fu₂, and n_e is the abundance of nematodes in these guilds, which represent an enriched condition of the food web (Ferris et al., 2001). Ba _{x} , Fu _{x} , Ca _{x} , Om _{x} , (where $x = 1$ – 5) represent the functional guilds of nematodes that are bacterivores, fungivores, carnivores and omnivores where the guilds have the characters indicated by x on the colonizer–persister (cp) scale (1–5) following Bongers and Bongers (1998).

2.5. Statistical analyses

Nematode abundance was log-transformed prior to statistical analysis for normality of data. We build generalized linear mixed models to test the main effect of treatments and their interactions. For a response variable y_{ijkl} (i represents replicates for CO₂ treatments, j represents treatments for CO₂, k represents treatments for nitrogen, l represents treatments for residual $i = 1, 2, 3$; $j = 1, 2$; $k = 1, 2$; $l = 1, 2, 3$). For normal distributed response variables

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