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The network of interactions among soil quality variables and nematodes: short-term responses to disturbances induced by chemical and organic disinfection

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ABSTRACT

Anthropogenic pressures that involve different kinds of inflows (chemical *vs* organic) to soils are likely to induce different responses of individual soil components as well as to affect the web of interactions between them. The aim of this paper was to explore whether chemical or organic disinfection constitute two different types of disturbance, regarding both their severity on individual soil components and the structural changes they induce on the network of interactions among soil biochemical variables and nematode functional guilds. Network analysis, a novel approach in the context of soil ecology, was used to explore these interactions.

Different plots within a field cultivated conventionally with *Phaseolus vulgaris* for many years were disinfected either with metham sodium or with a mixture of plant-based alternatives (neem and essential oils). Control plots receiving no disinfectants were also included in our study. One month after the treatments were applied, we estimated microbial C and N, activities of asparaginase, glutaminase, urease and phosphatase, organic C and N, inorganic N and P as well as the functional diversity of nematodes.

Chemical disinfection had a direct lethal effect on all nematodes, while the effect on the microbial community was less obvious, implying that at the time of sampling, microbial populations, especially bacterial ones had started to recover from disturbance. Urease and phosphatase activities were inhibited, which may partly be responsible for the reduced amounts of inorganic N and P. Organic disinfectants reduced microbial populations, mainly the fungal ones, but they did not inhibit enzyme activities. The availability of N and P increased and nematode abundance was not affected significantly.

The results demonstrate the usefulness of network analysis in providing insight into the structure and robustness of the soil network and its response to disturbance. Despite the pronounced reduction in nematode numbers due to chemical disinfection, the importance of nematode guilds within the interaction network was amplified and a more compact network was formed. On the other hand, interactions within organic plots were found to be primarily dictated by soil biochemistry. Finally, the analysis showed that both types of disinfection increased the vulnerability of the interaction network and this was more pronounced in chemically treated plots.

1. Introduction

Anthropogenic pressures on soils should be evaluated not only by the intensity with which soil components respond, but also by the changes induced in the web of interactions between them.

Disinfection with chemical fumigants falls within the strongest pressures on agricultural soils, suppressing soil borne pathogens (Welsh et al., 1998), nematodes (Giannakou et al., 2002), insects (Desaeger et al., 2004) and weeds (Gilreath and Santos, 2004), and affecting soil enzymes (Stromberger et al., 2005; Klose et al., 2006) and the process of C and N mineralization (Ibekwe et al., 2001; Collins et al., 2006). Among chemical disinfectants, metham sodium is one of the most commonly used (Pruett et al., 2001), due to its broad biocidal activity in the soil (Weiss and Lowit, 2004). Over the last two decades, there has been a growing interest in soil disinfection by means of non-chemical alternatives, such as plant-based products, which are considered to cause less severe disturbance to agro-ecosystems (Akhtar and Mahmood, 1994)

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and even to improve the soil physico-chemical properties (Gopal et al., 2007).

The aim of this paper is to explore whether chemical or organic disinfection constitute two different types of disturbance, inducing different responses in soil components. For this purpose, an experiment was conducted in a field cultivated conventionally with shell beans (*Phaseolus vulgaris* L.) for many years. Different plots within the field were disinfected either with metham sodium or with a mixture of plant-based alternatives. One month after treatments, we estimated microbial C and N, organic C and N, inorganic N and P, which are all considered to be linked with soil quality (Schloter et al., 2003), activities of four enzymes (L-asparaginase, L-glutaminase, urease and acid phosphatase) that control key metabolic pathways in soil and are used as an index of microbial diversity (Nannipieri et al., 2002; Klose et al., 2006), and all nematode functional guilds, due to their contribution to soil nutrient turnover (Ferris and Matute, 2003; Ferris et al., 2004).

Although quantitative estimation of the above mentioned biochemical and faunal parameters is informative regarding the intensity of the exerted pressures, it does not offer insights into the possible qualitative structural changes within the soil system, because the interactions among soil components are not considered. To explore how soil biochemical parameters and nematodes combine to produce an interaction total, we used network analysis techniques, which are suitable for the study of interaction webs of any kind (Hanneman and Riddle, 2005). A network is a collection of interacting nodes that may represent any level of the biological hierarchy such as chemicals, genes, proteins, organs, individuals, populations, etc. In our case, each node represents either a nematode functional guild or a biochemical variable. Ties between nodes may represent various interactions such as biochemical interactions, energy flows, competition coefficients in community studies, functional responses in predator–prey systems (Proulx et al., 2005). In our study, node ties represent correlation coefficients among abundances of nematode guilds and concentrations of biochemical variables. In ecology, relevant techniques have been employed by ecosystem ecologists to study input/output models of energy and/or material flows, such as food webs (e.g. Ulanowicz, 1987; Yodzis and Winemiller, 1999; Jordán et al., 2006). Within this framework, another task of this paper was to examine the extent to which network analysis might offer insights with respect to the structure and robustness of the soil network.

Our hypothesis was that chemical and organic disinfection would not only differ regarding the severity of their impact on individual soil components, but they would affect the structure of the soil network in different ways, because different kinds of inflows (chemical *vs* organic) are involved.

2. Materials and methods

2.1. Study site

The study was conducted in the Prespa region, located in the Northwestern part of Greece. The climate is mild continental-central European with Mediterranean features as well. The average annual precipitation ranges between 600 and 900 mm, and the wet season lasts from October to May. Snowfall is common from October until April (data from WWF-Greece, 2002). The soil of the experimental area consists of recent alluvial deposits derived from gneisses and granito-gneisses (Kosmas et al., 1997).

The soil physical properties of our study site (Table 1) were determined as described in Sections 2.2 and 2.3. The soil is characterized as sandy-loam with quite high values of bulk density. The latter however does not reach the value of 1.75, which is considered a limit for root growth (Arshad et al., 1996).

Table 1
Physical soil properties of the experimental site, estimated from $n = 9$ soil samples 20 cm in depth.

Physical variables	Mean ± SE
Silt (%)	25.16 ± 1.88
Clay (%)	18.83 ± 1.07
Sand (%)	56 ± 2.43
pH	5.9 ± 0.064
Bulk density (g cm^{-3})	1.63 ± 0.033

2.2. Experimental design and sampling

Within an area of 3000 m^2, which was conventionally cultivated with shell beans for several years, a field plot experiment was set up consisting of 18 plots (10 m × 10 m each). During the time interval between the cultivation periods of beans and, more specifically, two months before the new sowing of the year, six replicate plots were sprayed with metham sodium (10 l NEMASOL-51SL 100 l^{-1} H_2O 100 m^{-2}), six were disinfected with a mixture of organic additives, whereas the remaining six received no disinfectants and were used as controls.

Organic additives consisted of neem cake (a product of the neem tree *Azadirachta indica* A. Juss) and a mixture of plant essential oils. The products of the neem tree apart from their insecticidal (Koul, 2004), anti-microbial (Coventry and Allan, 2001) and nematicidal properties (Akhtar and Malik, 2000), act at the same time as an organic fertilizer that improves the soil physico-chemical properties (Gopal et al., 2007). The oil mixture (trade name: VIVERE-FYT) consisted of basil oil, marigold and clove oil, which are recommended in organic farming to be used together with neem (http://www.agrosector.gr) for their suppressive effects on soil microbes (Isman, 2000) and nematodes (Pandey et al., 2000). Neem was dispersed by hand (10 kg 100 m^{-2}) and immediately afterwards the oil mixture was sprayed over the plots (0.33 l 100 l^{-1} H_2O 100 m^{-2}). The quantities of both chemical and organic additives are those recommended by the manufacturers (Taminco N.V., Gent, Belgium for NEMASOL-51SL and GEOVET-HELLAS S.A., Veria Imathias, Greece for Neem-cake and VIVERE-FYT).

Surface tillage with a disc harrow followed the application of all disinfectants to incorporate them into the soil. Control plots were also tilled, so that the effect of tillage would not bias our results. All plots receiving different treatments were randomly interspersed following a randomized complete design (3 treatments × 6 replicate plots). Untreated 2 m wide strips were left between plots. Before setting up the field experiment, soil physical properties were estimated once from nine soil samples taken randomly within the experimental site by means of an auger 7.5 cm in diameter and 20 cm in depth.

To test the short-term effect of treatments on soil biochemical variables and nematodes, samplings were carried out one month after disinfection, first to allow enough time for the residual activity of metham sodium, which according to the manufacturers persists for 25 days, and second to avoid additional disturbance due to the beginning of the new cultivation period, i.e. deep tillage, sowing and fertilizer application. For the determination of soil biochemical variables, samples were collected from the upper 20 cm of the soil by an auger 7.5 cm in diameter. For the study of nematodes, each soil sample was a composite of three soil cores 3 cm in diameter and 20 cm in depth. In both cases, three soil subsamples were taken from each plot to account for within-plot heterogeneity.