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Use of pepper crop residues for the control of root-knot nematodes

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Abstract

The biofumigant effect of pepper crop residues (PCR) for controlling *Meloidogyne incognita* populations was evaluated. Under laboratory conditions, 0, 5, 10 and 20 g PCR were applied to 500 g nematode infested soil, with four replicates per treatment. After 20 days at 25 °C, PCR reduced significantly *M. incognita* populations and root galling indices in susceptible tomato cv. Marmande, and increased K, N and organic C in soil. In the field, biofumigation with PCR combined with fresh animal manures (with and without plastic cover), methyl bromide, and a control were evaluated through root galling indices on a pepper crop. Each treatment, except for the control, had a grafted and non-grafted susceptible pepper sub-treatment, with three replicates. Root galling indices were lower, and yields higher, on grafted plants, biofumigation with PCR and plastic cover, with similar values as MB treatment, suggesting that biofumigation with PCR is an efficient non-chemical alternative to control *M. incognita* populations, especially when applied with plastic cover, nitrogen-rich organic matter and followed by grafting on resistant pepper.

Keywords: Organic amendments; Biofumigation; Solarization; *Meloidogyne incognita*; Integrated crop management

1. Introduction

Root-knot nematodes (*Meloidogyne* spp.) cause major economic damage in agricultural production worldwide. The main options for control of phytoparasitic nematodes include chemical nematicides, crop rotation and resistant cultivars when available. The broad host spectrum of *Meloidogyne* species makes crop rotation difficult. Fumigant nematicides, although effective, have negative side effects that have led to their ban or restricted use. Resistance-breaking populations of *Meloidogyne* are challenging the use of resistant cultivars (Kaloshian et al., 1996; Peixoto et al., 1997; Castagnone-Sereno, 2002a,b; Robertson et al., 2006).

Organic amendment of soils has been used since the beginning of the agriculture. It benefits physical and chemical soil properties, increases soil fertility and aids in pest control (García Álvarez et al., 2004). Organic soil amendments used for nematode control are extremely heterogeneous, including green manures, animal manures, animal "beds" (sawdust, straw), composts, soil urban residues, and a variety of agro-industrial by-products (Cook and Baker, 1983; Hoitink, 1988; D'Addabbo, 1995). A variant of amending soil with organic material is biofumigation, a technique relying on the fumigant action of volatile compounds released during biodegradation for the control of plant pathogens (García Álvarez et al., 2004). It requires use of fresh, non-degraded organic material (Bello et al., 2002; Bello et al., 2003). It is also important to retain the gases that are produced for at least two weeks, because their effect is often biostatic, making it necessary to have an extended exposure time of the pathogen to the gases (García Álvarez et al., 2004).

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Both amendment of soil with organic matter and biofumigation were included as non-chemical alternatives for nematode control by the Methyl Bromide Technical Options Committee (MBTOC, 2002). Laboratory and field experiments have shown that biofumigation stimulates the biological activity of soils, increasing the populations of antagonistic organisms of pathogens such as *Pseudomonas fluorescens* and *Bacillus* spp., as well as the populations of saprophagous and bacteriophagous nematodes, and decreasing plant-parasitic fungi and nematodes. These results suggest the existence of a relationship between the biological activity of soils and pathogen suppressiveness (El Titi and Ipach, 1989; Werner and Dindal, 1990; Chen et al., 1991; Gamliel and Stapleton, 1993; Scow et al., 1994; Sarathchandra et al., 1996; Bello et al., 1997; Abawi and Widmer, 2000; Riegel and Noe, 2000; Chavarría-Carvajal et al., 2001).

Among the large variety of organic materials from animal and vegetable origin that have been tried as biofumigants, agricultural by-products, and especially crop residues, are increasingly becoming of interest. Although crop residues are usually considered as "wastes", materials without any value, and even as contamination sources, their incorporation into the soil contributes to the nutrient and organic matter recycling into the system, decreasing losses in organic matter and energy, as well as the costs needed to compensate those losses. Besides, crop residues secondary metabolites, which are generally produced inside plant tissues and released during the decomposition process, may be introduced into the soil (Gamliel et al., 2000). It was found that when crop by-products are used as biofumigant materials the compounds released are mainly aldehydes and isothiocyanates, which have biocidal activity and have been related to control of plant-parasitic nematodes (Sayre et al., 1965; Abawi and Widmer, 2000; García Álvarez et al., 2004).

Earlier, we found that agricultural and agro-industrial residues such as orange juice industry wastes, tomato, pepper, strawberry and cucumber crop by-products (alone and combined with sheep or commercial manure), showed promise for nematode control (Piedra Buena et al., 2006). As our interest is focused on crop by-products, we have performed new assays to confirm these results. The aim of this work was to evaluate the biofumigant effect of pepper crop by-products and establish the most efficient dose for the control of *Meloidogyne incognita*, as well as their effect on other edaphic organisms, on root galling of tomato and on soil fertility.

2. Methods

2.1. Laboratory assays

A loamy soil collected in pepper greenhouses from Campo de Cartagena (Murcia, Spain), with alkaline pH, high salinity, and high population levels of *M. incognita* (121 J2/100 g) was used for the biofumigation assays performed at the Department of Agroecoloy, CCMA-CSIC (Madrid, Spain). The assays followed a protocol developed by Bello et al. (2003), biofumigation treatments consisting of 0, 5, 10 and 20 g pepper crop residues (PCR) on 500 g soil, in a ratio of 4:4:1:1 for leaves:stems:petioles:fruits (fresh weight), with four replicates (plastic bags) per treatment. A dose of 5 g biofumigant material per 500 g soil is equivalent to 25 t/ha in field, which is the remaining biomass of a pepper crop after harvest (Bello et al., 2003).

The biofumigation assay was done over a 20-day-period at 25 (±1) °C in the dark. After this period, nematodes were extracted from 100 cm^3 soil of each replicate using sugar centrifugation (Nombela and Bello, 1983). The number of *M. incognita* juveniles (J2) dead and alive, rhabditids, dorylaimids and enchytraeids were counted at 40× magnification. A portion of 300 g soil of each replicate was put in pots, and a 15 day-old tomato seedling cv. Marmande (susceptible to *M. incognita*) was transplanted into each pot. The plants were kept in a growth chamber (24 ± 1 °C, 14 h daylight) for 33 days, when the soil was washed from the roots for visual evaluation of root galling (Bridge and Page, 1980). The remaining soil of each replicate was dried at room temperature (20 ± 2 °C) for one week, and ground in a mortar for chemical analysis (MAPA, 1994).

The data were statistically analyzed using ANOVA, and means of each treatment were compared with the Least Significant Difference (LSD) test ($p \leqslant 0.05$), using the SPSS statistical software for Windows (standard version 11.5.1, SPSS Inc., 2002).

2.2. Greenhouse experiments

The greenhouse biofumigation experiment was done over a three crop cycle period (3 years) at the IMIDA (Murcia, Spain) as a part of a multi-year trial on management practices to control soil nematodes and diseases of pepper. Data were collected during the third crop cycle. There were six treatments, with three replicates each. Individual plots were 3 × 18 m (54 m^2), with two pepper rows. One row was planted with pepper cv. Almuden grafted on pepper cv. Atlante (resistant to *M. incognita*, Ramiro Arnedo S.A.), and the other row with non-grafted pepper cv. Almuden. The control treatment had non-grafted plants only. A non-treated row was left between plots and not evaluated. Five plants per row (total number of plants per row = 45) were collected to evaluate root galling. The treatments were

- *Control*: Placed on the external rows of the greenhouse, these plots were not disinfected for two consecutive crop cycles, while in the third crop cycle (the one of the experiment presented) three different biological products were applied: Prosnema (Plymag, a mixture of 4.2% aminoacids + nematophagous fungi: *Arthrobotrys oligospora*, *A. dactiloides*, *Dactylella* spp., *Harposporium anguillulae*, *Mycrothecium*, etc.; De Liñán, 2005) at 10 $l\,ha^{-1}$ in one row, and Azadiractin (Sipcam Inagra,