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Detection and Description of Soils with Specific Nematode Suppressiveness

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Abstract: Soils with specific suppressiveness to plant-parasitic nematodes are of interest to define the mechanisms that regulate population density. Suppressive soils prevent nematodes from establishing and from causing disease, and they diminish disease severity after initial nematode damage in continuous culturing of a host. A range of non-specific and specific soil treatments, followed by infestation with a target nematode, have been employed to identify nematode-suppressive soils. Biocidal treatments, soil transfer tests, and baiting approaches together with observations of the plant-parasitic nematode in the root zone of susceptible host plants have improved the understanding of nematode-suppressive soils. Techniques to demonstrate specific soil suppressiveness against plant-parasitic nematodes are compared in this review. The overlap of studies on soil suppressiveness with recent advances in soil health and quality is briefly discussed. The emphasis is on methods (or criteria) used to detect and identify soils that maintain specific soil suppressiveness to plant-parasitic nematodes. While biocidal treatments can detect general and specific soil suppressiveness, soil transfer studies, by definition, apply only to specific soil suppressiveness. Finally, potential strategies to exploit suppressive soils are presented.

Key words: biological control, cyst nematodes, cyst nematode-suppressive soil, density dependence, heat treatments, *Heteordera avenae, H. glycines, H. schachtii.*

Soilborne pathogens and pests persist in a complex soil environment. Biotic and abiotic factors interact to influence disease development and survival of the microbial populations. Several articles, book chapters, and books have described microbial antagonists to plant-parasitic nematodes (Carris and Glawe, 1989; Carris et al., 1989; Chen and Chen, 2001; Jatala, 1986; Mankau, 1980; Meyer et al., 1990; Poinar and Jansson, 1988; Stirling, 1991; Tribe, 1977). A low level of biological buffering against plant diseases and pests is expected to be present in any agricultural soil and is called anti-pathogenic potential or general soil suppressiveness (Baker and Cook, 1974; Sikora, 1992; Weller et al., 2002). This type of suppressiveness is removed when the soil is sterilized. The occurrence and persistence of general soil suppressiveness may be reduced due to intensive agricultural practices. The continued detrimental loss of organic matter in some of the current production systems, e.g., intensive cotton production, is recognized (Entry et al., 1996) and might reduce the resilience to plant pathogens.

In contrast, the benefit for soil health by organic matter amendments and(or) crop sequences is proposed (Widmer et al., 2002). Soil health is the product of a number of ecological interactions (Herrick, 2000), and the improvement of these in agricultural soils is considered as the critical component of sustainable agricultural production (Doran and Zeiss, 2000). For example, amendments with organic matter increase microbial activity in soil resulting in improved soil health, which in turn is characterized by the increased resilience toward plant parasites and pathogens (Abawi and Widmer, 2000; Lazarovits, 2001; Van Bruggen and Semenov, 2000). To this end, soil suppressiveness is associated with soil health (Van Bruggen and Semenov, 2000). Mechanisms of specific suppression of fungal diseases have been reviewed (Mazzola, 2002; Weller et al., 2002). The usefulness of nematode-suppressive soils for studying biological control of plant-parasitic nematodes is widely accepted (Stirling, 1991). It is commonly believed that an improved exploitation of biological control mechanisms will greatly benefit from a thorough understanding of natural mechanisms that regulate nematode population densities. Nematode-suppressive soils, although poorly understood, often contain an array of nematode antagonistic microorganisms (Kerry, 1990).

Nematode-suppressive soils often are first recognized or suspected when population densities of the nematode decline after initial establishment (Gair et al., 1969) or when populations remain significantly lower in some fields than in other fields in the same area with similar soil and crop histories (Carris et al., 1989; Westphal and Becker, 1999). Suppressive soils often are associated with monoculture of a susceptible host (Gair et al., 1969; Hartwig, 1981; Heijbroek, 1983; Noel and Wax, 2003; Westphal and Becker, 1999). However, monoculture does not invariably lead to a nematode-suppressive soil (Carris et al., 1989). Field observations of suspected nematode-suppressive soils must be confirmed by greenhouse tests to determine the intrinsic character of the soil suppressiveness. In these tests, general soil suppressiveness is distinguished from specific suppressiveness by the fact that the latter is transferable. This review primarily covers the detection of the specific soil suppressiveness, defined by Baker and Cook (1974) for soilborne diseases as "the inhospitability of certain soils to some plant pathogens is such that either the pathogen cannot establish, they establish but

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fail to produce disease, or they establish and cause disease at first but diminish with continued culture of the crop." Methods to study specific soil suppressiveness will be discussed. Greenhouse tests have been developed and used to characterize suppressive soils for a number of soilborne diseases (Mazzola, 2002; Weller et al., 2002), and a number of similar methods can be used to identify nematode-suppressive soils (Kerry, 1988). Nematode-suppressive soils occur worldwide, but only a limited number of examples have been demonstrated to be biological in nature (Crump, 1989; Kerry, 1988). The biological nature of specific suppressiveness is confirmed when suppressiveness (i) is eliminated by biocidal treatments, (ii) can be transferred to conducive soil with small portions of suppressive soil, (iii) is specific to a particular pathogen (Kerry, 1988), (iv) can be observed as reduced reproduction in cyst and root knot nematodes in the root zone, (v) can be isolated by baiting techniques, (vi) is heat sensitive, and (vii) is density dependent (Westphal and Becker, 2001a,b,c). The objectives of this article are to review and summarize such techniques for identifying nematode-suppressive soil with particular reference to cyst nematodes, and to discuss limitations of the techniques as well as how suppressive soils can be manipulated. The emphasis is on a general description of suppressiveness with a view toward identifying the major factors involved in it. A perspective on currently available tools to manipulate suppressive soils is presented. The impact of facultative bacterial nematode antagonists is only briefly discussed in relation to naturally occurring suppressive soils.

Elimination of suppressiveness with biocidal treatments: Gair et al. (1969) followed population densities of *Heterodera avenae* and other plant-parasitic nematodes under cereal monoculture for several growing periods and found that population densities declined after initially high population densities. Typically, nematode population densities increased initially before declining to low levels (Kerry, 1987). Formaldehyde drenches of soils in which nematode decline had occurred and cropping of a susceptible host resulted in increased nematode reproduction in comparison to non-treated controls (Kerry et al., 1980; Williams, 1969). This preliminary observation of lower population densities in non-treated natural soil led to detailed studies of organisms contributing to nematode suppression and ultimately to the identification of *Nematophthora gynophila* and *Verticillium chlamydosporium* as microorganisms primarily responsible for maintaining nematode population densities below the damage threshold (Kerry et al., 1980, 1982a,b). Biocide treatments—some of them strictly experimental and others at pesticide label rates—have been used to reduce microbial populations and to reduce biological soil suppressiveness. Methyl bromide, methyl iodide, formaldehyde, metam sodium, treatments with aerated steam, micro-waving or autoclaving, followed by infestation with the plant-parasitic nematode have been used to demonstrate the biological nature of suppressive soil (Bird and Brisbane, 1988; Weibelzahl-Fulton et al., 1996; Westphal and Becker, 1999; Zuckerman et al., 1989). The nematode life stage used for infesting such treated soils is critical. For example, when egg suspensions were used to inoculate untreated soils, population densities of *Meloidogyne javanica* were suppressed compared to population densities in formaldehyde-treated equivalents of these soils, but suppression of nematode populations was limited when egg masses were used as inoculum (Orion et al., 2001). In these trials, a myriad of microorganisms capable of reducing nematode infectivity was associated with the single eggs (Orion et al., 2001). Organisms capable of consuming free eggs might not be specialized nematode parasites, and the inoculation with configurations of nematode life stages that are not present in agricultural soils, e.g., free root-knot nematode eggs, will probably be of limited help in detecting suppressive soil without further confirmation. Reduction of population densities in the undisturbed soil following inoculation with various life stages supports the claim for soil suppressiveness. For example, a California soil, infested with *H. schachtii* in 1975, supported only low numbers of the sugar beet cyst nematode under continuous cropping of host plants (Fig. 1) (Westphal, 1998). Biocide treatments of this soil with various compounds followed by inoculation with either cysts or J2 demonstrated the biological nature of this soil (Table 1) (Westphal and Becker, 1999).

When using biocide treatments to test for nematode parasites or predators alike, possible side effects of the biocides and the conditions under which they are tested must be considered. Non-treated controls often exhibit poor root growth when field soils are placed in greenhouse pots. Thus, a reduction in the number of feeding sites could confound the detection of nematode suppression (Crump and Kerry, 1987; Westphal and Becker, 1999). In cyst nematodes, the ratio of eggs per cyst indicates whether the nematodes reproduce freely. The ratio of eggs and (or) cysts per root weight will be a measure of nematode suppression independently of the availability of feeding sites. Although biocides are typically non-specific, their direct effects on microorganisms are often associated with alterations in the physical and (or) chemical properties of the soil in ways that could differentially influence the nematode and the plant (Sandler et al., 1988). If the application of various materials with the common denominator of biocidal activity but varying side effects results in similar removal of soil suppressiveness, the claim for biological suppressiveness is supported. Then, effects of these materials on soil chemistry and physics seem of minor concern.

Transferability of suppressiveness with small portions of soil: In soilborne diseases, specific suppressiveness (e.g.,

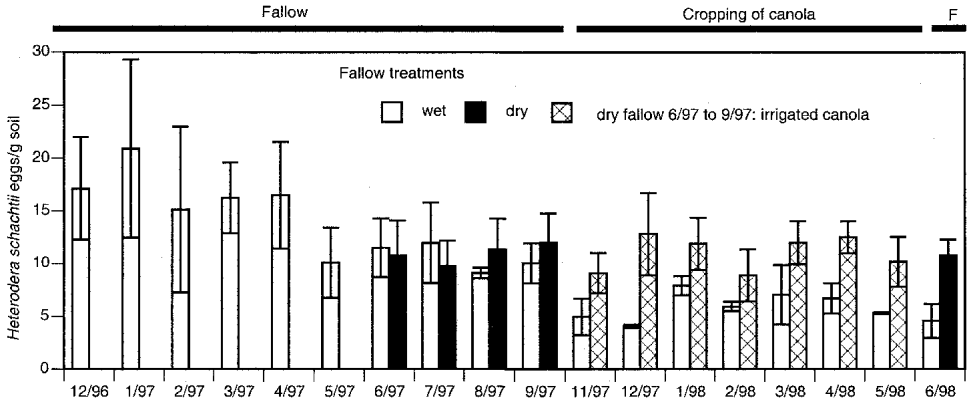

FIG. 1. *Heterodera schachtii* population densities during December 1996 until June 1998 in field 9E under moist and dry fallow in 1997 and consecutive cropping with rapeseed. Bars represent standard error. Soil samples were taken from the upper 10-cm soil layer from the root zone of rapeseed when applicable (Westphal, 1998).

potato scab and take-all decline of wheat) is transferable to conducive soil with small portions of soil, and this observation was considered an indicator of the biological nature of suppressiveness (Menzies, 1959; Shipton et al., 1973). Transferability of suppressiveness is an indication of specific soil suppressiveness against plant-parasitic nematodes (Kerry, 1988), particularly when the nematode antagonists are not culturable or are unknown. Amendment of steam-sterilized greenhouse soil with *Pasteuria penetrans*-infested soil resulted in suppression of *Meloidogyne incognita* by the obligate nematode parasite (Mankau, 1975). Transfer of the 20–53-um fraction of a soil derived from northern Europe that contained *Nematophthora gynophila* to South Australia soils infested with *H. avenae* resulted in fungal infection of the nematodes (Stirling and Kerry, 1983). This soil transfer approach is especially helpful when the active organism(s) is not yet identified. For example, soil transferability demonstrated the biological nature for *Criconomella xenoplax*-suppressive peach orchard soil when 5% of non-steamed orchard soil was mixed into the steamed peach orchard soil (Kluepfel et al., 1993). In *H. schachtii*-suppressive soil, such transfer was achieved in a field trial with 1% and 10% suppressive soil and in the greenhouse with as little as 0.1% suppressive soil to conducive soil (Westphal and Becker, 2000). In the field study, onset of soil suppressiveness was monitored with a bioassay for infective J2 in field plots. Suppressiveness increased in the 10% transfer treatment in initially conducive plots more rapidly than in the 1%-transfer treatment. It also was indistinguishable from the suppressive control after a shorter incubation in the higher—rather than in the lower—soil amendment treatment. This observation provided additional evidence of the biological nature of the nematode suppression (Fig. 2). When diluting test soil, less root rot and corresponding loss of feeding sites will occur; but when diluting a suppressive soil, effects on nematode reproduction will still be measurable. For example, in *H. schachtii*-suppressive soil, egg numbers per cyst were approximately 40, whereas they were almost 120 in conducive soil (Westphal and Becker, 2000).

Specificity of suppressiveness toward the pathogen: Biological control organisms are considered host specific com-

TABLE 1. Effect of biocidal treatments of nematode-suppressive soil on plant growth, *Heterodera schachtii* population density, and frequency of second-stage juvenile invasion of roots in greenhouse experiments[a] (Westphal and Becker, 1999).

	Greenhouse experiments					Bioassays	
Preplant treatment[b]	Top dry weight (g)	Root dry weight (g)	Cysts/g soil	Eggs/g soil	Eggs/cyst	J2/cm root length	J2 invaded (%)[c]
Control	3.8 c	0.4 d	0.5 c	21.2 d	34.2 c	0.6 c	0.1 c
Metam sodium	7.5 b	4.6 b	1.3 ab	220.3 ab	166.8 a	59.1 b	0.7 b
Methyl bromide	8.8 a	5.2 ab	1.3 ab	202.3 abc	151.1 ab	94.7 ab	0.9 b
Methyl iodide	9.1 a	6.0 a	1.3 ab	181.7 bc	136.2 b	96.1 ab	1.0 b
Formaldehyde	6.9 b	3.5 c	1.5 a	255.1 a	152.2 ab	57.3 b	0.8 b
Aerated steam	9.7 a	5.5 ab	1.1 b	150.7 c	131.8 b	118.9 a	1.5 a
LSD, $P = 0.05$	1.2	0.3	0.3	61.2	29.8	41.8	0.4
P for treatment F	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

[a] For each column, treatment means followed by the same letter were not significantly different compared using Fisher's LSD at $P = 0.05$. Data from two tests were combined for statistical analysis.

[b] The application rates were equivalents as follows: metam sodium 356 kg a.i./ha; methyl bromide 335 kg a.i./ha; methyl iodide 503 kg a.i./ha; formaldehyde (37% a.i.) 3,000 L/ha; aerated steam 30 minutes at 87.5 °C.

[c] Percentage of *H. schachtii* second-stage juvenile detected per centimeter root length of radish in relation to the number of *H. schachtii* eggs present in the 60-g soil sample.

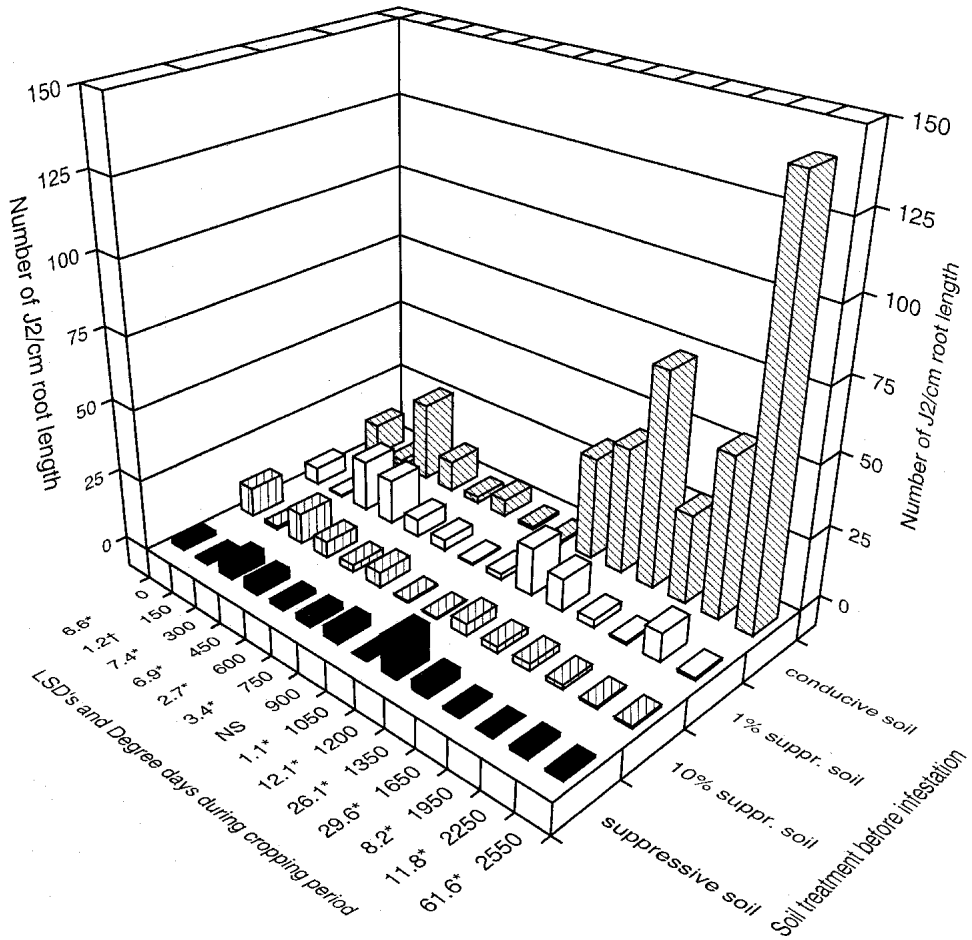

FIG. 2. Infectivity of *Heterodera schachtii* after four different soil treatments under Swiss chard at 150- or 300-DD intervals. The non-infested, fumigated control and the non-infested, untreated control were included in the statistical analysis but are not shown. The LSD values are indicated for * $P = 0.05$ and † $P = 0.10$ level (Westphal and Becker, 2000).

pared to chemical or physical control tools. Whereas some nematode antagonists can suppress plant-parasitic nematodes with similar life histories, a suppressive soil may function against a number of plant-parasitic nematodes. Specialized interactions of plant-parasitic nematodes and their antagonists have been reported. *Pasteuria* spp. for example, generally have a limited host range, but some strains can also infect nematodes other than the original host (Chen and Dickson, 1998). Some fungi, such as certain *Fusarium* spp. *Verticillium* spp., and the obligate prokaryote *P. penetrans,* have host ranges that include both cyst and (or) several root knot nematodes (Davies et al., 2001; Godoy et al., 1982; Meyer et al. 1990; Qadri and Saleh, 1990). Soil suppressiveness against *H. schachtii* was effective against *M. incognita* (Pyrowolakis et al., 2002). In these tests, soil suppressiveness was eliminated by soil fumigation. Such tests are valuable for characterizing soil suppressiveness in greater detail because components of soil suppressiveness that are active against particular life stages of a nematode may impact similar life stages of other nematodes.

Observation chambers to monitor population dynamics in situ: Various observation chambers have been used to study nematode-suppressive soils. Various types of straight and slanted containers have been used for nematode detection (Behringer, 1967; Sikora et al., 1985). Containers with at least one transparent side are filled with test soil and planted to a susceptible host; nematode females of cyst nematodes on the roots are observed through the transparent surface. Such chambers were particularly helpful for monitoring population dynamics when nematode population development in the chambers follows the same pattern as in field soil (Fig. 3) (Crump, 1987; Crump and Kerry, 1977; Westphal and Becker, 2001a). Containers with a triangular base and three transparent sides also allow the investigator to monitor development of nematode population density and to collect individual nematode specimens at specific time intervals (Crump, 1987). The alternative process of bulk extraction of intact cysts from soil risks losing nematode egg and cyst parasites that destroy the integrity of the nematode cyst. In general, use of observation chambers allows non-destructive observation of the root surface and permits investigation of processes in the root zone when nema-

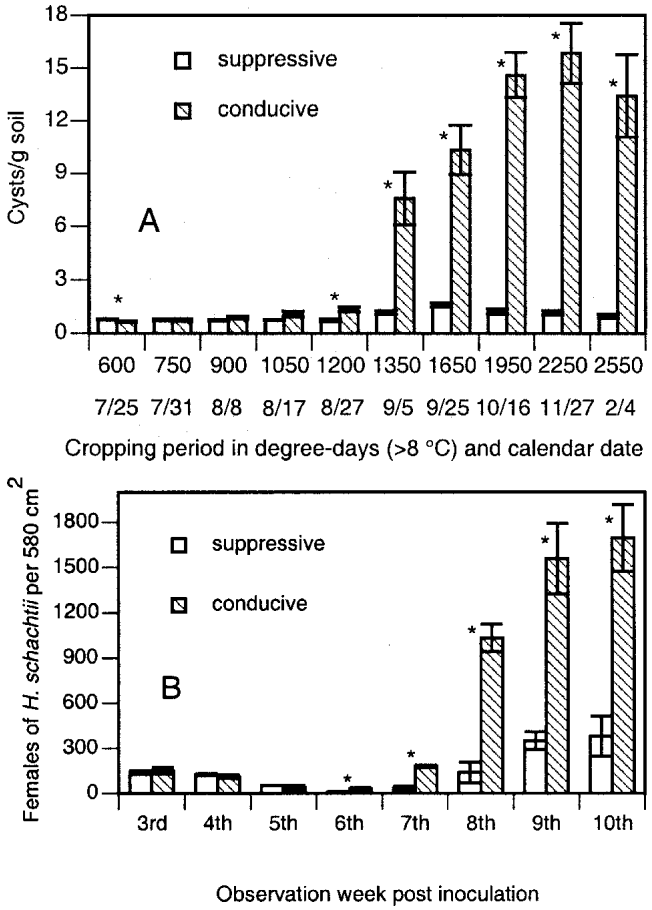

FIG. 3. *Heterodera schachtii* population densities in suppressive and conducive soils. A) Cyst population densities during 230 days (2,550 degree-days, base temperature 8 °C) under Swiss chard in field plots. B) Female population densities on mustard-greens roots in root observation chambers. * Significant differences according to Fisher's protected LSD at $P = 0.05$. Bars represent standard error (Westphal and Becker, 2001a).

tode population dynamics otherwise could not be determined. For example, the aggressive antagonist *N. gynophila* was detected only because its activity was observed in the root zone of cereal (Crump and Kerry, 1977). The researchers observed cyst nematode development and subsequent destruction by the parasites. The additional benefit of these chambers is the ability to recover nematode specimens of a known age and with a known life history; such nematodes can then be examined further and used for additional tests, e.g., transfer studies (Westphal and Becker, 2001a).

Baiting and transfer with nematode life stages: Different nematode life stages have been proposed as transmitters of nematode suppressiveness. Although Nicolay and Sikora (1989) found that cysts were not the main site of reproduction of fungal nematode parasites, nematode cysts were recently demonstrated to be efficient transmitters of nematode suppression when cysts that had developed in a suppressive soil induced suppressiveness in a conducive greenhouse soil at the rate of 1 cyst per 110 g of soil (Table 2) (Westphal and Becker, 2001a). This approach narrowed the range of organisms that need to be tested further, but both nematode parasites or organisms that exhibit nematode antagonism could be discovered this way. This transferability with cysts allowed the analysis of microbial populations associated with cysts by culture-independent strategies (Yin et al., 2003a,b). Whereas the cyst transfer approach helped identify the target of nematode antagonists, it might be less suitable for detecting nematode suppression because field populations of nematode cysts often are small and may be broken and non-recoverable during extraction. In strict baiting approaches, healthy nematode life stages of the target nematode are added to test soils in different carrier systems to improve recovery, or they are added directly to the test soil and re-extracted. The closer the nematode life stage and its configuration in egg masses or intact cysts are to those in the environment, the more meaningful are such bioassays. When a sugar beet cyst nematode-suppressive soil was tested for suppressiveness against *M. incognita,* suppression was comparable after inoculation with nematode juveniles or egg suspensions, supporting the hypothesis that this soil was suppressive against both nematodes (Pyrowolakis et al., 2002). Cysts of *H. schachtii* can be embedded between layers of nylon cloth, framed, and inserted into test soils; after a 2-week incubation in greenhouse pots, nematode cysts are recovered, broken open, and the

TABLE 2. Suppression of *Heterodera schachtii* by amendment with cysts from different sources in comparison with non-amended infested soil[a] (Westphal and Becker, 2001a).

Transfer source	Experiment 1			Experiment 2		
	Root dry weight	Cysts/g soil	Eggs/cyst	Root dry weight	Cysts/g soil	Eggs/cyst
Non-amended	3.5	1.8 a	51.2	1.5	4.2 a	51.7
Cysts, conducive soil (rootbox)	3.1	1.7 ab	52.7	1.9	3.9 a	63.5
Cysts, suppressive soil (rootbox)	3.0	1.2 c	52.6	2.2	2.8 b	34.5
Cysts, 1% suppressive field 9E soil	3.3	1.4 bc	76.6	2.2	2.4 b	43.3
1% suppressive field 9E soil	2.9	1.4 c	66.9	1.9	2.7 b	44.9
LSD $P = 0.05$	N.S.	0.3	N.S.	N.S.	0.50	N.S.
P for treatment F	0.4888	0.0017	0.2743	0.1833	0.0001	0.1605

[a] The amendment cysts were collected from suppressive field soil or from the corresponding root observation chambers. Treatments with the same letter were not significantly different when tested with Fisher's protected LSD at $P = 0.05$.

egg content stained to facilitate counting of fungal-infected nematode eggs (Kiewnick and Sikora, 1994).

In another baiting technique, surface-sterilized nematode eggs are embedded in an alginate film carried by a nylon screen (Rodríguez-Kábana et al., 1994). Although both techniques have limitations, baiting with cysts reflects natural conditions in the soil more closely than single egg exposure. In the field, a biocontrol organism needs to gain access to eggs, embedded in nematode cysts or egg masses. Thus, the use of entire cysts seems appropriate. In contrast, single eggs of root-knot nematode were subjected to attack by soil organisms, whereas egg masses were partially protected from microbial infection by the gelatinous matrix (Orion et al., 2001).

In yet another baiting method, healthy nematode juveniles are added to test soil, re-extracted after varying times of incubation, and examined for infection for surveying for known nematode parasites. With such techniques, a survey for root-knot nematode suppression was conducted in South Australia (Stirling and White, 1982). In that study, three procedures were used to find the distribution and importance of the target nematode antagonist *P. penetrans*. Nematode specimens were observed directly as collected, or nematode migratory stages were extracted, or soils were inoculated with baiting nematodes, which were incubated for various times in the soil and then recovered and examined for infection (Stirling and White, 1982). It is critical to test for infection rather than attachment alone because some *Pasteuria* spp. can attach to nematode cuticles but not infect (Davies et al., 1990). These attaching, but not infecting species are likely of limited value as biocontrol organisms. Baiting techniques have the common advantage of narrowing the range of organisms that must be studied to identify mechanisms in a suppressive soil. The closer to the natural conditions the baiting material is delivered to the test, the more likely effective candidate organisms will be isolated. To provide the entire life history in the test soil for baiting, host plants were planted in field soils in greenhouse pots and nematode specimens collected from the roots after incubation and used for isolating an unidentified fungus, ARF 18 (Kim et al., 1998).

Heat sensitivity of soil suppressiveness: Heat treatments are commonly used to reduce soil microbial communities in substrates for container-grown plants (Baker and Roistacher, 1957). Aerated steam is typically used to apply selective heat treatments, which are effective because microorganisms have different lethal temperatures. The steam-air mixture provides an efficient delivery system for heat units; varying temperatures are delivered by adjusting the ratio of steam and air. For example, selective heat treatments were helpful in elucidating the contributors to suppressiveness against Fusarium wilt (Rouxel et al., 1977). Other examples of removal of soil suppressiveness by soil pasteurization have been reported (Kluepfel et al., 1993; Westphal and Becker, 1999). In a recent study, heat was applied by submerging the test soil in plastic bags in a waterbath at a range of preset temperatures (Westphal and Becker, 2001a). This treatment selectively eliminated fractions of the microbial populations that otherwise confer suppressiveness in the natural non-treated soil (Table 3) (Westphal and Becker, 2001a). Elimination by selective heat is another indication that soil suppressiveness is biological in nature. Elimination of selected groups of microorganisms helps narrow the organism groups most important in the soil suppressiveness. The concomitant elimination of root-rot organisms presents a methodological problem similar to non-specific biocidal treatments in that the improvement of root health also results in more nematode feeding sites in treatments that eliminate nematode suppression.

Density dependence of suppressive soil: Density dependence is an important characteristic of the interaction of biological control organisms and soilborne pests and pathogens (Jaffee, 1993). This phenomenon is an indicator of the intimate interaction of the nematode pest and its antagonists. Density dependence was demonstrated, for example, in the interaction of *H. schachtii* and the nematode-pathogen *Hirsutella rhosiliensis* (Jaffee et al., 1992, 1993). When effects of soil suppressiveness were examined for various soilborne fungal dis-

TABLE 3. Effect of heat treatment of *Heterodera schachtii*-suppressive soil on microbial groups and consecutive *Heterodera schachtii* population development (range: 45 °C to 65 °C)[a] (Westphal and Becker, 2001a).

	Precropping/posttreating monitoring			Harvest weights		*H. schachtii* populations		
Heat treatment	Pseudomonas spp. ($\times 10^4$/g soil)	*Pythium* spp. (CFU/g soil)	F. oxysporum ($\times 10^3$/g soil)	top dry (g)	root dry (g)	cysts/ g soil	eggs/ g soil	eggs/ cyst
Control	9.5 a	226.7 a	3.8 a	3.7 cd	0.3 b	1.0 b	65.4 b	76.4 b
45 °C	5.1 a	222.2 a	2.9 b	3.3 d	0.5 b	0.8 b	85.0 b	86.9 b
50 °C	0.0 b	6.7 b	1.4 c	5.0 bcd	0.8 b	2.1 b	218.5 b	95.7 b
55 °C	0.0 b	0.0 b	0.0 d	5.9 abc	1.7 a	5.3 a	831.7 a	157.0 a
60 °C	0.0 b	0.0 b	0.0 d	6.3 ab	1.8 a	5.6 a	836.6 a	166.9 a
65 °C	0.0 b	1.1 b	0.0 d	7.5 a	2.2 a	4.6 a	961.9 a	184.4 a
LSD $P = 0.05$	4.9	39.9	0.6	2.2	0.6	1.4	327.5	40.5
P for treatment F	0.0018	0.0001	0.0001	0.0149	0.0001	0.0001	0.0001	0.0001

[a] Treatments with the same letter within one column were not significantly different when tested with Fisher's protected LSD at $P = 0.05$.

eases, typically, disease onset occurred at lower inoculum levels and disease symptoms were expressed sooner in conducive than in suppressive soils (Burke, 1965; Louvet et al., 1981; Smith and Snyder, 1971). Only a few nematode-suppressive soils have been studied for density dependence. In the study of a *H. schachtii*-suppressive soil in southern California, soil suppressiveness was density dependent. This interaction was measurable with a radish bioassay and at harvest of a field crop of Swiss chard grown in the microplots 2 months after infestation of field microplots and incubation at approximately half of the field capacity (Fig. 4) (Westphal and Becker, 2001b). Even without knowing the antagonists responsible for soil suppressiveness, this kind of study described the density-dependent interaction of the nematode with the suppressing principals in soil.

Management of plant-parasitic nematodes is only one aspect of managing sustainable productivity of agricultural soils. Nematode-suppressiveness in soils is a fascinating biological phenomenon in continuous flux. Whereas short-term observations suggest a steady state of these soils from the standpoint of sustained low nematode population densities (Westphal, 1998), long-term observations have detected fluctuations in nematode population densities (Heijbroek, 1983), as expected for a host-parasite interaction. Exploitation of soil suppressiveness for nematode management has been difficult because such soils are often induced under non-practical monoculture with susceptible host plants and the resulting initially high population densities of the plant-parasites. Recent evidence suggests that suppressiveness can also develop under resistant host plants (Noel and Wax, 2003), a possibility that was not considered after the failure of *N. gynophila* and *V. chlamydosporium* to increase under resistant host crops in *H. avenae*-infested soil (Kerry and Anderson, 1984). Soil suppressiveness against *H. schachtii* was maintained under resistant oilseed radish or sugar beet but was lost under wheat, a non-host plant (Westphal and Becker, 2001c). These findings should change the thinking in nematode management in that crop sequences must be considered with a focus on effects on soil suppressiveness rather than the pest populations per se.

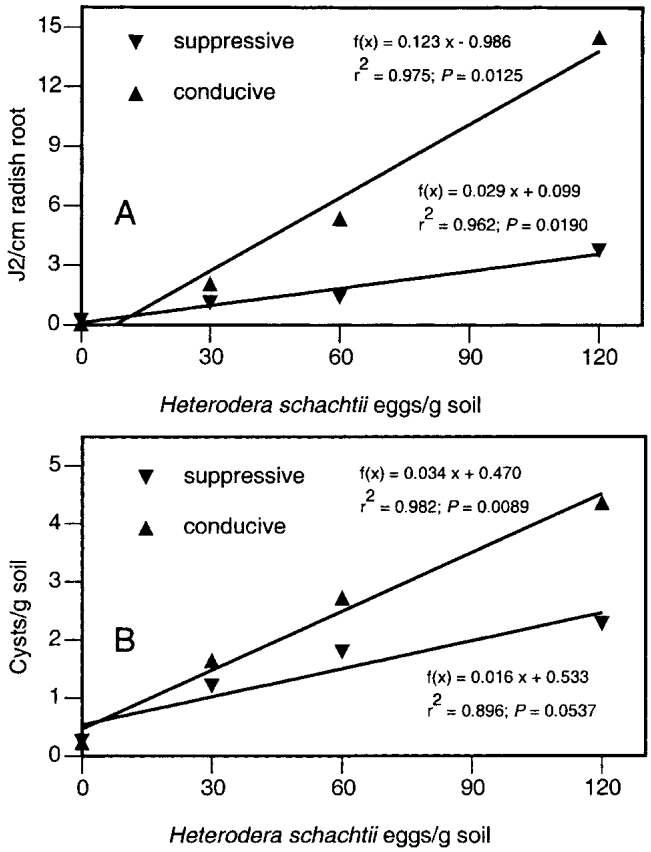

FIG. 4. *Heterodera schachtii* population densities in suppressive and conducive soil in field microplots infested with greenhouse-raised cyst nematodes at the equivalent of the egg populations indicated with linear regression. A) At planting penetration by juveniles of radish roots in suppressive soil infested at different egg densities of *H. schachtii* in a growth chamber bioassay 2 months before planting. B) Cyst population densities in suppressive and conducive soil at harvest of Swiss chard (Westphal and Becker, 2001b).

Inundative approaches of amendment of nematode-conducive soils with nematode parasites or predators have common difficulties in that effectiveness is either dependent on large amounts of organic matter or has experimental challenges in demonstrating nematode suppression (Kerry, 1990). While some inundative approaches of controlled release of nematode antagonists have been attempted (Chen and Dickson, 1998; Jaffee, 2000; Stirling et al. 1998ab), procedures on how to manipulate the soil microbial communities in an integrated approach to induce soil suppressiveness or enhance the biological antagonistic potential are necessary. Pioneering work of Linford et al. (1938) used this approach in the 1930s by adding large amounts of organic matter to nematode-infested soil to enhance biological suppression. Many soil amendments of crop or animal wastes have been proposed, and chitin-containing materials have received particular attention (Abawi and Widmer, 2000; Culbreath et al., 1985; Muller and Gooch, 1982; Rodríguez-Kábana, 1986; Rodríguez-Kábana et al., 1984). These approaches require excessive quantities of organic material, which makes them impractical for intensive, large-scale agriculture. Recent work has tested the possibility of providing such amounts of organic matter by cover-cropping with nematode antagonistic plants (Hoffmann-Hergarten and Sikora, 1992; Pyrowolakis et al., 1999). One report proposes the use of nematicides to assist establishing suppressive soils (Fernandez et al., 2001). This is an exciting research area that nematologists and soil ecologists who are interested in soil health and quality can investigate together (Abawi and Widmer, 2000; Van Bruggen and Semenov, 2000).

Tillage practices can influence nematode population dynamics as well. Some reports indicate that no-tillage

practices reduced soybean cyst nematode in certain crop sequences (Edwards et al., 1988; Noel and Wax, 2003). Differences of tillage effects on nematode population densities were measurable mainly in finely textured soils (Workneh et al., 1999), but it is not known whether these nematode population density changes are associated with changes of microbial communities. It will be challenging to elucidate the mode of action that suppresses the soybean cyst nematode under no-tillage. Recent work on soilborne plant-pathogenic fungi has demonstrated an exciting novel approach: only one cultivar of wheat grown as cover crop induced suppressiveness against apple replant disease by increasing disease-suppressive Pseudomonads (Mazzola and Gu, 2002), exemplifying that substantial information is still to be discovered.

The understanding of soil microbial communities is only marginal when considering the much lower frequencies of culturable microorganisms compared to non-culturable frequencies that determine different aspects of diversity (Hill et al., 2000). Novel techniques for studying soil microbial communities are being developed (Hill et al., 2000) but have been used only sparingly for the study of nematode-suppressive soils (Yin et al., 2003a,b). Future applications of such novel approaches that consider entire cropping systems when trying to induce soil suppressiveness are promising and deserve more attention. Exploitation of soil suppressiveness is a long way from reality, but the phenomenon is worth investigating, particularly as we strive for sustainable agriculture while the reduced availability of nematicides and soil fumigants places current production systems at risk.

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