

## Manipulation of Rhizosphere Bacterial Communities to Induce Suppressive Soils

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**Abstract:** Naturally occurring disease-suppressive soils have been documented in a variety of cropping systems, and in many instances the biological attributes contributing to suppressiveness have been identified. While these studies have often yielded an understanding of operative mechanisms leading to the suppressive state, significant difficulty has been realized in the transfer of this knowledge into achieving effective field-level disease control. Early efforts focused on the inundative application of individual or mixtures of microbial strains recovered from these systems and known to function in specific soil suppressiveness. However, the introduction of biological agents into non-native soil ecosystems typically yielded inconsistent levels of disease control. Of late, greater emphasis has been placed on manipulation of the cropping system to manage resident beneficial rhizosphere microorganisms as a means to suppress soilborne plant pathogens. One such strategy is the cropping of specific plant species or genotypes or the application of soil amendments with the goal of selectively enhancing disease-suppressive rhizobacteria communities. This approach has been utilized in a system attempting to employ biological elements resident to orchard ecosystems as a means to control the biologically complex phenomenon termed apple replant disease. Cropping of wheat in apple orchard soils prior to re-planting the site to apple provided control of the fungal pathogen *Rhizoctonia solani* AG-5. Disease control was elicited in a wheat cultivar-specific manner and functioned through transformation of the fluorescent pseudomonad population colonizing the rhizosphere of apple. Wheat cultivars that induced disease suppression enhanced populations of specific fluorescent pseudomonad genotypes with antagonistic activity toward *R. solani* AG-5, but cultivars that did not elicit a disease-suppressive soil did not modify the antagonistic capacity of this bacterial community. Alternatively, brassicaceae seed meal amendments were utilized to develop soil suppressiveness toward *R. solani*. Suppression of *Rhizoctonia* root rot in response to seed meal amendment required the activity of the resident soil microbiota and was associated with elevated populations of *Streptomyces* spp. recovered from the apple rhizosphere. Application of individual *Streptomyces* spp. to soil systems provided control of *R. solani* to a level and in a manner equivalent to that obtained with the seed meal amendment. These and other examples suggest that management of resident plant-beneficial rhizobacteria may be a viable method for control of specific soilborne plant pathogens.

**Key words:** suppressive soils, biological control, replant disease, rhizobacteria.

The study of naturally occurring disease-suppressive soils has produced significant progress towards acquiring an understanding of the biotic and abiotic forces that inhibit plant disease development in such soils (Weller et al., 2002). Identification of the functional biological entities that contribute to the observed disease suppression has provided a wealth of microbial resources with potential to be utilized as agents for the biological control of soilborne plant pathogens. Most commonly, these organisms have been evaluated for the capacity to perform as disease control agents when introduced into cropping systems as single or multiple strain inoculants (Cook, 1993). Decades of research in a multiplicity of cropping systems have utilized an inundative release model at the time of planting as the means to incorporate microorganisms, such as plant growth-promoting rhizobacteria, into soils with the goal of attaining disease control. In most instances, such attempts in field-level commercial production systems have failed to attain the perceived promise of these resources for control of soilborne diseases. Such an out-

come should not come as a surprise, as the survival and activity of any microorganism in an alien biological system while in competition with the myriad of microbes adapted to that same soil would not be expected. Likewise, disease suppression in any given soil may require the interaction among multiple biotic and abiotic factors, and the function of a specific agent, at sites external to that specific system, could effectively be diminished.

Recently, more realistic approaches have been evaluated in attempts to achieve biologically mediated soilborne disease management. Rather than inoculating soils or plant propagative materials with mass produced formulations of non-native biological agents, an emerging strategy has employed practices to manage the biology resident to the soil system with the goal of inducing soil suppressiveness. Such a management process would be expected to require some knowledge of the functional microbial populations that are contributing to the observed disease suppression. In some instances, knowledge exists of the specific biological agent(s) responsible for the development of a disease suppressive soil, with a few of the more prominent examples being those soils suppressive to take-all of wheat, potato scab and *Fusarium* wilt of various crop plants (Weller et al., 2002). Given knowledge of the functional biological mechanism, a real opportunity exists to utilize agro-

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nommic practices as a means to manage the resident microbial community in a manner leading to a reduction in disease.

However, for the vast majority of soilborne diseases, although a suppressive soil may have been identified, studies are still necessary to elucidate the operative mechanism(s) prior to attempting manipulation of the biological system. Unfortunately, attempts to manage soil suppressiveness through methods such as the addition of organic amendments have commonly been undertaken without this essential biological knowledge. Such studies most likely will fail to yield the capacity to predict whether disease suppression will be attained by a particular practice, thus usefulness of the findings are limited to the specific soil or medium in which the experiments were conducted.

An on-going research program at the USDA-ARS Tree Fruit Research Lab seeks to harbor the activity of biological resources native to orchard ecosystems as a means to control apple replant disease (Mazzola, 1998). One element of this program has attempted to utilize the resources endemic to the production system, whether this encompasses the plant itself or other biological features interacting with the host, as a means to control this disease. This paper will endeavor to describe our attempts to manage specific elements of the resident rhizobacteria community for the control of soilborne plant pathogens, with particular emphasis on work which has sought to manage *Rhizoctonia solani*, one element of the pathogen complex that incites apple replant disease.

#### SUPPRESSIVE SOILS

Soils suppressive to soilborne plant diseases have been defined as those in which disease development is minimal even in the presence of a virulent pathogen and susceptible plant host. As noted above, examination of naturally occurring disease-suppressive soils has yielded significant information as to the biological traits that lead to disease control and provided insight into possible means to manage the phenomenon. Such soils and the corresponding attributes contributing to disease suppression have been described for numerous plant diseases including those incited by *Heterodera* spp. (Kerry, 1988; Westphal and Becker, 1999; Yin et al., 2003a, 2003b), *Streptomyces scabies* (Menzies, 1959), *Fusarium oxysporum* (Stotzky and Martin, 1963; Scher and Baker, 1980), *Gaeumannomyces graminis* var. *tritici* (Cook and Rovira, 1976), *Phytophthora cinnamomi* (Broadbent and Baker, 1974), *Plasmodiophora brassicae* (Murakami et al., 2000), *Pythium* spp. (Hancock, 1977) and *Rhizoctonia solani* (Henis et al., 1978, 1979).

Although certain suppressive soils are naturally occurring and are dependent upon physical or chemical attributes of the soil, in other systems the capacity of a soil to limit disease progression develops over time in

response to specific management systems (Weller et al., 2002). Perhaps the most notable illustration of the latter model is soils suppressive to take-all of wheat, which is incited by *G. graminis* var. *tritici*. During the initial years of continuous wheat monoculture, disease incidence increases but a spontaneous decline in disease severity, termed take-all decline, is realized after an indeterminate period of time, and soil remains suppressive as long as wheat monoculture is not interrupted (Gerlagh, 1968; Shipton et al., 1973). The fact that the phenomenon is observed across geographic regions, is elicited by the same biological factor across soils and occurs in response to a specific agronomic practice (Weller et al., 2002; De Souza et al., 2003) is considerable evidence to indicate that resident microbial resource management holds promise as an effective soilborne disease control strategy.

#### MANAGEMENT OF RESIDENT SOIL MICROBIAL COMMUNITIES TO INDUCE DISEASE SUPPRESSION

Although the biologically functional components of disease-suppressive soils are well documented for certain systems, there has been an absence of success in transforming this knowledge into effective disease management programs. Such an outcome is influenced by several factors, ranging from the perception that such a goal would be too difficult to obtain, to the lack of interest in moving from a program of biological discovery to one of an applied field-based nature. However, there are sufficient small-scale successes achieved in such environments as container-based production systems (Boehm and Hoitink, 1992) to warrant our attempts to induce soil suppressiveness on a field-based production system.

Management of resident bacterial communities to induce disease suppression can take several forms, but from a practical standpoint it is limited to a minimal number of options. In the case of take-all of wheat, it is known that a certain threshold population of 2,4-diacetylphloroglucinol (2,4-DAPG)-producing fluorescent pseudomonads must be attained to achieve a suppressive soil (Raaijmakers and Weller, 1998) and that certain genotypes of these bacteria are more effective in suppressing disease than others. Thus, identification of a wheat genotype with a greater capacity to enhance resident populations of 2,4-DAPG-producing pseudomonads or a more active genotype of these bacteria might allow for more rapid development of take-all decline. In fact, wheat cultivars do differ significantly in their ability to support resident populations of 2,4-DAPG in the rhizosphere and appear to exhibit varying preferences for genotypes of these bacteria (Mazzola et al., 2004). For most systems, there is substantially less information available concerning the functional elements of the soil system that elicit suppression of a particular soilborne disease. As such, additional back-

ground trials are required that provide a broader examination of the biological system and not simply a determination of what plant cultivar to employ. In general, attempts to foster soil suppressiveness have focused on use of specific cropping systems or specific soil amendments to enhance the activity of the functional biological community and thus will be the focus of the following discussion.

#### MANIPULATION OF RHIZOSPHERE BACTERIAL COMMUNITY THROUGH PLANT CROPPING SYSTEM

Modification of the cropping system has long been employed as a means to control soilborne plant diseases. The most common and effective scheme has been the use of crop rotation, with disease control achieved as the absence of a suitable plant host results in diminished viability of the pathogen. Attempts to develop specific cropping models to manage the resident soil microflora for disease suppression have been few. It has been reasoned that as increased plant diversity can enhance microbial community biomass (Zak et al., 2003), mixed cropping systems will generate a more diverse microbial community and thus should be more resilient to pathogen invasion (Workneh and van Bruggen, 1994; Hiddink et al., 2005). However, the preponderance of examples of induced-suppressive soils come from crop monoculture systems (Chet and Baker, 1980; Cook and Weller, 1987), and limited attempts to compare mixed opposed to single crop systems indicate that mixed systems may not enhance microbial diversity or disease suppressiveness (Hiddink et al., 2005).

In attempting to develop an integrated approach for the control of apple replant disease, the use of a pre-plant cover crop was explored as a potential component of such a system. The goal of employing this tactic was to modify the resident microbial community in a manner that induced soil suppressiveness to one element of the causal pathogen complex, *R. solani* (Mazzola, 1998). Wheat was the chosen cover crop, as it was observed that soil collected from a field formerly cropped to continuous wheat monoculture was suppressive to disease incited by the apple pathogen *R. solani* AG-5 but immediately adjacent ground that recently had been converted to apple was conducive to disease development (Mazzola, 1999). It was noted that orchard soils became increasingly conducive to *R. solani* as age of the orchard block from which soils were collected increased. The decrease in soil suppressiveness corresponded with a precipitous decline in rhizosphere populations of *Burkholderia cepacia* and *Pseudomonas putida*. In greenhouse trials, application of successive short-term wheat cropping sequences to soils enhanced growth of apple seedlings subsequently planted into these orchard soils (Mazzola and Gu, 2000). Enhanced seedling growth was associated with a reduction in root infestation by *R. solani* and *Pratylenchus penetrans*, as well

as significant increases in rhizosphere populations of fluorescent pseudomonads and a transformation in composition of the population to one dominated by *P. putida* (Mazzola and Gu, 2000).

These initial studies were followed by a more detailed analysis of wheat cropping effects on fluorescent pseudomonad populations and plant root infection by *R. solani*. Orchard soils were initially conducive to the development of Rhizoctonia root rot of apple and wheat, incited by an introduced isolate of *R. solani* AG-5 and AG-8, respectively. Prior to wheat cultivation, the fluorescent pseudomonad population resident to these orchard soils was dominated by a genotype that did not inhibit in vitro growth of *R. solani*. Wheat cropping of orchard soils resulted in the induction of soil suppressiveness, which developed over a period of three successive 28-day wheat crop cycles and occurred in a wheat cultivar-specific manner (Mazzola and Gu, 2002). A transformation in genetic composition of the fluorescent pseudomonad community was observed, and specific genotypes were enhanced in the rhizosphere of wheat cultivars that induced soil suppressiveness. In vitro assays demonstrated that a greater proportion of fluorescent pseudomonad genotypes recovered from the rhizosphere of wheat cultivars that induced soil suppressiveness exhibited greater antagonistic activity toward *R. solani* than did the population from wheat cultivars that did not elicit a disease suppressive soil (Mazzola and Gu 2002; Gu and Mazzola, 2003). The fluorescent pseudomonad population recovered from the rhizosphere of apple planted in wheat-cultivated orchard soils exhibited a parallel conversion (Gu and Mazzola, 2003). Genotype 1, as designated by RFLP analysis of the 16S rRNA gene, dominated the population recovered from the rhizosphere of wheat or apple when grown in the non-treated control soil and those orchard soils previously cropped with the wheat cultivar Eltan, Hill-81 or Madsen. Isolates of this genotype do not inhibit *R. solani* (Table 1). In contrast, though genotype 1 was well represented in the population recovered from the rhizosphere of apple or wheat planted into Lewjain or Penewawa cropped soils, this genotype represented a significantly smaller proportion of the total fluorescent pseudomonad population, and at least 40% of the isolates recovered exhibited a high level of antagonism toward *R. solani*.

A similar wheat cropping system was applied at an orchard site during the year prior to re-planting apple. This field trial employed three wheat cropping sequences of approximately 10 weeks in duration over the course of a single growing season, with the wheat shoot biomass removed prior to planting the subsequent wheat crop. The wheat cropping treatment was as effective as pre-plant soil fumigation with methyl bromide in reducing *Rhizoctonia* spp. infection of Gala/M26 apple roots at this site and significantly improved tree growth and yield (Mazzola and Mullinix, 2005). How-

TABLE 1. Impact of wheat cultivation on proportional distribution of 16S rDNA genotypes among fluorescent *Pseudomonas* spp. populations recovered from the rhizosphere of different wheat cultivars, or apple seedlings grown in wheat cultivated orchard soil.

16S rDNA genotype <sup>z</sup>	In vitro antagonism of <i>R. solani</i> AG-t <sup>y</sup>	Control		'Eltan'		'Hill-81'		'Lewjain'		'Madsen'		'Penewawa'	
		W <sup>x</sup>	A	W	A	W	A	W	A	W	A	W	A
1	0	78.3 <sup>w</sup>	68.3	70.0	58.3	73.3	58.3	41.7	36.7	75.0	58.3	48.3	46.7
2	+++			5.0	3.3	5.0	1.7	15.0	3.3	8.3	5.0	25.0	30.0
4	+											1.7	
6	++		1.7			1.7	1.7			5.0			
9	+		5.0		3.3	1.7	3.3		15.0				8.3
10	0		3.3	3.3	3.3	1.7	15.0	1.7				10.0	5.0
11	+++		1.7	3.3		10.0		8.3	5.0	3.3	3.3	6.7	1.7
13	+	10.0	1.7	5.0	3.3	1.7	6.7	5.0	11.7	3.3			3.3
14	0		1.7				3.3			3.3			
21	+++				1.7				1.7				
27	++								13.3		3.3		1.7
29	0				3.3		6.7				3.3		1.7
30	0						3.3		3.3				
33	0	1.7	10.0	5.0	6.7								
34	0				8.3						3.3		
35	0	3.3		10.0		1.7		8.3					
38	+++	6.7				3.3		18.3		1.7		8.3	

<sup>z</sup> Genotypes were defined by RFLP analysis of an amplified fragment of 16S rDNA.

<sup>y</sup> In vitro antagonism was defined by the zone of inhibition (mm) between expanding hyphal growth of *R. solani* AG-5 and the margin of the bacterial colony when grown on nutrient broth-yeast extract agar. 0 = no zone of inhibition; + = >0 to 4 mm; ++ = >4–7.9 mm; +++ = >8 mm.

<sup>x</sup> W = wheat; A = Apple.

<sup>w</sup> Values are a percent of the total population recovered from an individual crop species, and each 'population' consisted of 60 individual fluorescent *Pseudomonas* spp. isolates.

ever, apple replant disease is incited by a consortium of pathogens (Mazzola, 1998), some of which were not controlled by this practice, and pre-plant fumigation proved superior to wheat cropping in improving overall apple growth and yield.

#### SOIL AMENDMENTS FOR INDUCTION OF SOIL SUPPRESSIVENESS

A wealth of studies have explored the application of various organic residues for the management of soil-borne plant diseases. Composts have been the most frequently used substrate in this context and have demonstrated significant levels of efficacy, particularly in controlled environment or container-based production systems (Hoitink et al., 1977; Mandelbaum and Hader, 1990; Widmer et al., 1998). In certain instances, the mechanism by which compost amendment provides control of a specific soilborne pathogen has been examined and determined to be of a biological nature (Boehm et al., 1997; Hoitink and Boehm, 1999). Most commonly, efficacy has been attributed to an overall increase in biological activity in the soil system, but in other systems a defined element of the microbial community and an operative mechanism have been established. A major limitation to this approach is the inability to predictably reproduce compost composition, both from a substrate and microbiological perspective. Utilization in field-level production systems further has been limited by the reported need to apply composts at rates of 5–20% (vol/vol) or higher to achieve disease

control (Noble and Coventry, 2005) even in relatively non-complex environments such as containerized production systems.

An alternative approach to the introduction of off-site materials has been the cultivation and incorporation of specific green manure crops into soil as a means to manage specific biologically active resident soil microbial communities. A green manure crop of buckwheat or canola increased the proportion of streptomycetes in the resident population that were antagonistic toward the potato pathogens *Streptomyces scabies*, *Verticillium dahliae* and *R. solani* (Wiggins and Kinkel, 2005a). The relative increase in inhibitory activity of the streptomycete community was frequently associated with a decrease in disease development and an increase in potato yields. Similar increases in the proportion of antagonistic streptomycetes and reduction in alfalfa root rot were observed in buckwheat or sorghum-sudangrass-treated soils (Wiggins and Kinkel, 2005b).

A recent development in utilization of bio-based amendments for disease control has been the application of defined substrates to shift microbial communities in a manner leading to disease suppression. Although a somewhat untested strategy, it has met with modest success. A wide range of studies have investigated brassicaceae plant residues for the suppression of various plant pests including pathogens, insects and weeds (Brown and Morra, 1997). However, the vast majority of such investigations have been undertaken with the notion that the degree of pest suppression is altogether a function of the loosely termed process "biofu-

migation” (Matthiessen and Kirkegaard, 2006). As plants within the brassicaceae produce glucosinolates, which upon hydrolysis yield various biologically active compounds including isothiocyanates, such a concept is compelling (Brown and Morra, 1997). An alternative to this presumed mechanism of action has been attained in studies examining the use of brassicaceae seed meals for the control of soilborne plant pathogens, most specifically *R. solani*. Several lines of evidence, discussed below, demonstrate a partial role or the absence of a role for the glucosinolate hydrolysis in the brassicaceae seed meal-elicited control of this fungal pathogen and implicate specific elements of the resident soil microbial community in mediating the observed disease control.

Initial studies were conducted to examine the impact of *Brassica napus* seed meal on root infection of apple incited by *R. solani* AG-5 (Mazzola et al., 2001). These experiments included a high (113  $\mu\text{mole/g}$ ) and low (21.3  $\mu\text{mole/g}$ ) glucosinolate content seed meal, both of which were sourced from the same cultivar, ‘Dwarf Essex’. Interestingly, regardless of the rate at which the seed meal was applied to soil, both provided an equivalent level of disease control (Mazzola et al., 2001). Subsequent studies demonstrated that a native soil microbial community was required to elicit the disease control response. Pasteurization of *B. napus* seed meal did not impact the level of disease suppression attained, but pasteurization of soil prior to seed meal amendment and infestation with inoculum of *R. solani* abolished capacity of the seed meal to suppress apple seedling root infection by this pathogen (Cohen et al., 2005). This same response to soil pasteurization was observed with multiple brassicaceae seed meals, including that derived from *Brassica juncea*, which is known to yield hydrolysis products (allyl isothiocyanate; AITC) that are active against *R. solani* (Mazzola et al., 2007). In the case of *B. juncea* seed meal, the mechanism of action was found to vary in a temporal manner, i.e., when the pathogen was added to pasteurized soil within 24 hours of seed meal amendment, effective disease control was attained. When addition of *R. solani* inoculum was delayed until 48 hours post-seed meal amendment, pathogen suppression in native or pasteurized soil was not observed. The pattern of observed disease suppression corresponded with the pattern of AITC generation, a process that was completed within 24 hours of seed meal amendment (Mazzola et al., 2007). Disease suppression was restored to native soils when incubated for a period of 4 weeks, and the re-establishment of disease suppression was associated with the elevation of resident *Streptomyces* spp. populations (Mazzola et al., 2007).

Although these studies demonstrated the need for an active resident microbial community to enable seed meal-elicited disease control, the operative element(s)

of this community and mode of action were not apparent. A survey of broadly defined microbial groups in seed meal-amended soils revealed that streptomycete populations were consistently elevated one to two orders of magnitude higher than in non-treated soil or soil treated with a seed meal that did not provide control of *R. solani* (Cohen et al., 2005). In field studies, numbers of *Streptomyces* spp. recovered from *B. napus* seed meal-amended orchard soil remained an order of magnitude higher than the non-treated control 18 months after application of the amendment. Numbers of *Streptomyces* spp. recovered from the rhizosphere of various apple rootstocks 6 months after planting were consistently higher when grown in brassicaceae seed meal-amended soils (Table 2).

Subsequently, studies demonstrated that *Streptomyces* isolates recovered from the rhizosphere of apple grown in seed meal-amended soils were capable of providing control of *R. solani* AG-5 in a manner similar to that observed for brassicaceae seed meals. Apple root infection by *R. solani* AG-5 was suppressed in split-root assays where a portion of the root system was cultivated in *B. napus* seed meal-amended soil and the remainder grown in soil infested with the pathogen but lacking seed meal amendment (Cohen et al., 2005). This response indicated that disease control observed is a plant-mediated response, likely functioning through an induction of plant defense responses. *Streptomyces* spp. isolates suppressed *R. solani* root infection in a fashion similar to that observed in response to seed meal amendment. Bacterial strains limited *R. solani* root infection under both single-container and split-root plant propagation systems and did so to a level that was equivalent to *B. napus* seed meal amendment (Cohen and Mazzola, 2006). These data indicate that control of *R. solani* in response to brassicaceae seed meal amendment in large part is elicited through amplification of populations and activity of the *Streptomyces* spp. community resident to the rhizosphere of plants grown in amended soils.

*Streptomyces* spp. produce a diversity of secondary metabolites possessing anti-microbial activity, and application of antagonistic streptomycetes has resulted in con-

TABLE 2. Effect of brassicaceae seed meal amendments on *Streptomyces* spp. populations (log cfu/g root) recovered from the rhizosphere of apple rootstocks grown in GC orchard soil.

Soil treatment	Apple rootstock			
	Seedling	G16	M7	M26
Control	4.15a <sup>z</sup>	4.09a	4.20a	4.20a
Pasteurization	3.77a	4.74ab	4.83ab	4.08a
<i>Brassica juncea</i>	5.65b	5.36bc	5.67c	5.63b
<i>Brassica napus</i>	5.53b	5.57c	5.86c	6.15b
<i>Sinapis alba</i>	4.27a	5.31bc	5.52bc	5.55b

<sup>z</sup> Means in the same column followed by the same letter are not significantly ( $P > 0.05$ ) different.

trol of numerous soilborne plant diseases (Jones and Samac, 1996; Chamberlin and Crawford, 1999; Samac and Kinkel, 2001). Although *Streptomyces* soil and rhizosphere populations increased in response to seed meal amendment, the proportion of isolates recovered from the apple rhizosphere demonstrating antagonism toward *R. solani* AG-5 was not altered (Cohen et al., 2005). The proportion of the population recovered from soil expressing antagonism toward *R. solani* AG-5 increased significantly, but there was no corresponding reduction in hyphal growth of this fungus through seed meal-amended soil (Cohen et al., 2005). Examination of a limited number of *Streptomyces* isolates detected no association between in vitro antagonism toward *R. solani* and capacity of an isolate to control root infection by this pathogen (Cohen and Mazzola, 2006). Taken together, these data suggest that induction of plant defense responses rather than antibiosis may be the driving mechanism by which resident streptomycetes elicit the seed meal-induced control of *R. solani*.

Brassicaceae seed meal amendment has proven to be effective in limited field trials for the control of *R. solani* infection of apple roots (Mazzola and Mullinix, 2005). These trials also demonstrate the need to recognize the pathogen-specific nature of biologically mediated "soil suppressiveness." Though *B. napus* seed meal amendment effectively controlled *R. solani*, the same treatment resulted in elevated populations of *Pythium* spp. As a result, use of this seed meal for the control of apple replant disease required a post-plant application of mefenoxam to suppress root infection by the complex of *Pythium* spp. resident to orchard ecosystems capable of parasitizing apple (Mazzola et al., 2002). Such a practice is not compatible with organic systems, and an alternative seed meal formulation was generated for use in such systems. The addition of *B. juncea* seed meal to the amendment suppressed the *B. napus*-induced elevation of *Pythium* spp. populations and apple root infection in greenhouse (Mazzola et al., 2007) and field trials (Table 3).

### CONCLUSIONS

The management of resident soil microbial communities for the control of soilborne diseases appears to be a logical progression in the utilization of knowledge

TABLE 3. Effect of a composite brassicaceae seed meal amendment on recovery of *Pythium* spp. from soils at the RF organic orchard, Chelan, WA.

Treatment	<i>Pythium</i> spp. cfu/g soil
Control	154.2b <sup>z</sup>
Telone-C17 soil fumigation	212.5b
<i>B. juncea</i> / <i>B. napus</i> seed meal <sup>y</sup>	45.8a

<sup>z</sup> Means in the same column followed by the same letter are not significantly ( $P > 0.05$ ) different.

<sup>y</sup> Composite seed meal consisted of a 1:1 mixture of *B. juncea* cv. Pacific Gold and *B. napus* cv. Dwarf Essex applied in the tree row at a rate of 8.5 m tonne/ha.

that has emanated from the study of naturally occurring disease-suppressive soils. However, a circuitous route has been taken prior to the recent examination of such a strategy as a viable disease control option. Decades of research focused on the identification and isolation of biological elements that contribute to the suppressiveness of soil to individual soilborne plant diseases. Subsequent efforts centered on elucidating the specific microbial traits responsible for pathogen suppression, the genetic regulation of such traits and optimizing conditions for production and release of individual microbial agents into the agro-ecosystem. While such an approach can have value in biologically uncomplicated systems, such as those utilizing a defined plant growth medium, e.g., rockwool, from an ecological perspective it is counterintuitive to expect the same strategy to succeed in a field-based production model across a diversity of soil systems.

The examination of disease-suppressive soils and the processes that lead to such states demonstrate the potential for the management of resident rhizosphere bacterial communities as a means to control soilborne diseases. In several instances, commonly employed agronomic practices such as crop monoculture were found to lead to a biologically mediated disease-suppressive status, and this state was maintained for the duration of the period in which the practice was utilized. Certainly the capacity to generate soil suppressiveness without a concerted effort to do so would support the case for the active management of resident microbial communities as a viable disease control strategy.

Studies reported here demonstrate that management of resident bacterial communities colonizing the plant rhizosphere can lead to effective control of specific soilborne diseases. Modification or enhancement of the disease-suppressive rhizobacterial community was attained via the use of cropping or amendment-based approaches. However, these studies also demonstrated certain qualifications and limitations to the use of such methods. Unexpectedly, the induction of soil suppressiveness to *R. solani* in response to wheat cropping of orchard soil was documented in a wheat cultivar-specific manner. Of greater concern was the observation that, depending upon the source plant species, brassicaceae seed meal amendment, while inducing soil suppressiveness toward *R. solani*, led to significantly elevated populations of *Pythium* spp. populations and apple root infection.

As is known to be true for naturally occurring disease-suppressive soils, the management of a specific component of the resident rhizobacteria population as a means to induce soil suppressiveness is likely to confer disease control only to a limited component of the pathogen population indigenous to a plant production system. Field-based systems commonly encounter a myriad of potential soilborne pathogens, and it is the

rare case where only a single pathogen requires control to achieve optimal plant production. In the application of strategies for inducing soil suppressiveness toward *R. solani* described above, it was apparent that within the context of controlling apple replant disease such a practice could only be utilized within the framework of an integrated pest management system. As a stand-alone control measure, the cropping or amendment-based manipulation of the rhizobacteria community effectively suppressed *R. solani* but failed to control or exacerbated other components of the disease complex. While this strategy shows promise and effectiveness, these studies again reveal the need for comprehensive studies of the soil ecosystem, rather than compartmentalized examination of such systems, if sustainable methods for soilborne disease control are to be attained.

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