Biological Control to Reduce Inoculum of the Tan Spot Pathogen *Pyrenophora tritici-repentis* in Surface-borne Residues of Wheat Fields

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ABSTRACT

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Field plots of conservation-tillage wheat straw naturally infested with *Pyrenophora tritici-repentis* were treated with bran-based inoculum of three potential biocontrol fungi, and reduction in the number of residue-borne pseudothecia (primary inoculum of the pathogen) was determined. *Limonomyces roseipellis* significantly reduced residue-borne primary inoculum of *P. tritici-repentis* in 3 of 4 yr of experiments; an unidentified agonomycete was effective in two of three experiments, but *Laetisaria arvalis* was ineffective. Although *L. roseipellis* reduced inoculum by 60–80%, greater reductions would be necessary for acceptable control of this disease. Retention of dry bran-based inoculum on straw was enhanced by the use of alginate, but further improvements in formulation and application methods are required for effective biocontrol of pathogens in surface-borne crop residues.

Conservation tillage is a farming practice in which crop residue is retained on the soil surface between cropping seasons to reduce soil erosion and conserve soil moisture. This beneficial practice would be used more widely if certain pest constraints could be overcome. Weeds and diseases can prevent successful use of conservation tillage, particularly in crops grown without rotation. In continuous, conservation-tillage wheat in the central plains of North America, *Pyrenophora tritici-repentis* (Died.) Drechs., the causal agent of tan spot, is a major residue-borne pathogen. The pathogen survives saprophytically in surface-borne wheat straw and produces pseudothecia and ascospores, the primary inoculum for tan spot epidemics that can cause losses as high as 49% (11). Despite the fact that secondary cycles of the disease are caused by wind-borne conidia, tan spot severity has been associated with the local concentration of primary inoculum in conservation-tillage residue (11,12). Further, the amount of primary inoculum in the field (number of pseudothecia per square meter) is correlated with tan spot severity (1,17) and yield loss (1). Biological control to reduce primary inoculum is a possible management approach for this and other residue-associated diseases.

Several previous studies suggested that there is a potential for microbial antagonism to *P. tritici-repentis* in infested residue. A laboratory study by Summerell and Burgess (15) showed that *P. tritici-repentis* survives poorly in residue that is in intimate contact with nonsterile soil. Persistence of the pathogen in infested straw was correlated with the composition of the microbial community in various microenvironments in a no-till field (9). A later study (18) indicated that prolonged periods of moisture occur in those microenvironments in which the survival and pseudothecial production of *P. tritici-repentis* is poorest. However, prolonged high moisture per se is not detrimental to *P. tritici-repentis* in the absence of other organisms (6,14). Taken together, these observations suggest that high moisture in wheat crop residue may hinder the activity of *P. tritici-repentis* by favoring the activity of antagonistic microflora. Indeed, we isolated several straw-associated fungi, including *Limonomyces roseipellis* Stalpers & Loerakker and an unidentified agonomycete capable of inhibiting growth and pseudothecial production by *P. tritici-repentis* in dead wheat leaves under wet conditions (≥ -0.1 MPa) (7).

In an earlier community analysis of microflora on straw (9), we found that *L. roseipellis* (which we designated "unidentified basidiomycete") and the agonomycete isolate "Sterile II" were characteristic members of microbial community types in which *P. tritici-repentis* was present at reduced levels. Further, these two chitinolytic fungi, *L. roseipellis* in particular, were among the earliest secondary saprophytes to invade surface-borne straw. *L. roseipellis* is a fast-growing fungus that suppresses straw-borne *P. tritici-repentis* under laboratory conditions (4,5), particularly at moderately warm temperatures and high-moisture levels. Although *L. roseipellis* is a pathogen of turfgrass, the pink patch disease it causes is usually not severe in mowed turfgrass: Many leaves in an affected patch are undamaged, and the growth rate of the grass is not reduced significantly (13). The fungus caused very little detectable damage to turfs of perennial ryegrass or creeping red fescue in Kansas, even at high-inoculum levels (W. F. Pfender, W. Zhang, and A. Nus, *unpublished data*).

Biological control of pathogens in surface-borne crop residues has received little attention. Much biological control research has focused on protection of the infection court, whether root, seed, or foliage (2). In these plant-associated microenvironments, the flux of plant-produced nutrients has a strong influence on the activity and interactions of microorganisms. This ongoing input of nutrients is absent in crop residues, in which nutrients decrease and become more refractile over time. There are numerous cases of antagonistic interactions between residue-borne phytopathogens and microbes in the soil, and a number of these have been exploited for biological control (2). However, the soil environment differs markedly from the microenvironment of surface-borne residues, notably in the more variable and extreme physical aspects of the latter and, consequently, in its microbiota. As a result, biological control in surface-borne crop residues presents some unique ecological problems for antagonists. There are also formidable technical challenges in formulating biocontrol agents and applying them to this habitat.

In this study, our objectives were to reduce the primary inoculum of *P. tritici-repentis* in surface-borne straw through biocontrol under field conditions and to begin to address practical problems of formulating biocontrol agents and applying them to crop residues.

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MATERIALS AND METHODS

Field tests for biological control of residue-borne *P. tritici-repentis* were conducted by applying inoculum of the biocontrol agents to the straw in a reduced-tillage wheat field after harvest of the tan spot-infected crop. Effectiveness of biocontrol was assessed by determining the number of pseudothecia of *P. tritici-repentis* per unit weight of straw several months after treatment. The field tests were performed during 4 yr, and several methods of inoculation were used.

In each year of the study, hard red winter wheat cv. TAM 105 was planted during late September at the Rocky Ford Experimental Farm near Manhattan, KS. Plots were infested during early November by applying oat kernels colonized by *P. tritici-repentis* (17) to the surface of the field at a rate of approximately 50 g/m^2. Irrigation was applied periodically to favor tan spot development. Wheat was harvested during late June or early July with a combine having no straw chopper or spreader. Immediately after harvest, the standing stubble and cut straw were chopped with a tractor-drawn rotary mower set at 5 cm above the soil surface; this chopped straw was redistributed evenly over the plot area before plot boundaries were marked, and biocontrol treatments were applied.

Inoculum preparation and application. Several fungi were tested as possible biocontrol agents. Basidiomycete *L. roseipellis* (isolate 3T163) and agonomycete Sterile II had been isolated from wheat straw. *Laetisaria arvalis* Burdsall was not originally isolated from straw but has been reported to antagonize several soil-associated fungi (3). These three potential biocontrol agents were antagonistic to growth and pseudothecial production of *P. tritici-repentis* under favorable moisture conditions in a laboratory study (7). *Pithomyces chartarum* (Berk. & M.A. Curtis) M.B. Ellis also was used in the field studies as a negative-check treatment; it failed to show antagonism to *P. tritici-repentis* in previous laboratory studies. All fungi were stored in lyophilized form and were transferred to clarified V8-juice agar medium (16) from storage at the beginning of each year's inoculum preparation procedure.

In 1987, inoculum of *L. roseipellis* was produced by growing the fungus at 24 C for 10 days in covered pans containing a twice-autoclaved mixture of wheat bran, millet seed, and water (3:2:4 by volume). The moist inoculum was chopped in a blender for several seconds at low speed, then taken immediately to the field for application. Each biocontrol plot (1.3 m^2) was sprinkled with 600 cc of inoculum. The field was irrigated lightly immediately after inoculum application.

In 1988, inocula of *L. roseipellis, P. chartarum,* and Sterile II were grown for 8 days at 24 C in the above-described bran/millet seed mixture. Inoculum was blended with water (600 ml of water per 280 cc of inoculum) for 10 s at low speed. This inoculum slurry was held on ice until applied to the field plots (after no more than 3 h). Slurry (700 ml, produced from 280 cc of inoculum) was applied per 0.9-m^2 plot by a hand-held powder sprayer (H. D. Hudson Co., Chicago, IL) modified by enlarging the openings on the interior portion of the outflow spout. The field was irrigated as described for 1987.

In 1989, inoculum was produced by growing the fungi on wheat bran that had been ground in a Wiley mill to pass through a 20-mesh sieve (1.0-mm openings), was mixed 1:2 (wt/wt) with water, and was autoclaved twice on successive days. The fermentations were done in aluminum-foil pans (18 × 18 cm) each containing 40 g (dry weight) of bran. Small blocks of agar-cultured fungus were placed at 2.5-cm intervals in a grid pattern on the autoclaved bran and incubated for 4 days at 24 C. This incubation was sufficient for the fungus to thoroughly colonize the bran, which was then diced, air-dried, and ground in a blender (without water) to pass through a 20-mesh sieve. To apply the dry inoculum to the plots of straw, the following method was used: Water was first sprayed onto the straw at a rate of 92 L/ha. The dry bran was dusted onto the plots by a hand-held sulfur-duster (Armitsu Baby, ENCAP Products Co., Mt. Prospect, IL) at a rate of 17 g/m^2 of plot. The dry bran was followed by 92 L/ha of 3% alginate (Kelgin low viscosity, Kelco, Chicago, IL) applied by an air-assist sprayer (10) and then 92 L/ha of 0.4 M $CaCl_2$ to gel the alginate. The field was irrigated lightly immediately after treatment.

Inoculum was produced in 1990 using the 1989 methods. The application method was similar, except that the initial water application was omitted; the sequence was 3% alginate (92 L/ha), dry-bran inoculum (40 g/1.8 m^2 = 222 kg/ha), and 0.4 M $CaCl_2$ (92 L/ha). The final field irrigation was omitted in 1990.

During each year, the check plots received no inoculum or carrier treatment and thus, provided data for pseudothecial production by *P. tritici-repentis* on straw that received only normal agronomic treatment. The plots treated with *P. chartarum* served as negative biocontrol-treatment checks, by which any effects on *P. tritici-repentis* pseudothecial production resulting from the effect of fungus-colonized bran could be detected.

Weeds within plots were controlled by hand pulling. Hot weather (air temperatures over 38 C) occurred periodically in July during every test year.

Field-plot design and treatment dates. For the first year's experiment, wheat was harvested on 8 July (1987), and biocontrol inoculum was applied on 10 July. The plots were 1 × 1.3 m and were arranged in a completely randomized design (CRD) with four replications.

In 1988, wheat was harvested on 16 June. Straw was raked from the plots on 28 June and stored indoors until use. On 21 July the straw was replaced on the plots, and biocontrol inoculum was applied. The plots, 1.3 × 0.7 m, were arranged in randomized complete blocks (RCB) with three replicate blocks.

The harvest date in 1989 was 26 June, and biocontrol inoculum was applied on 5 July. Plots were 1 × 1 m, and were arranged in an RCB design with four replicate blocks.

In 1990, wheat was harvested on 22 June. Biocontrol inoculum was applied on 25 June in an RCB design with four replicate blocks. Plots were 1.3 × 1.3 m.

In all experiments, individual plots were separated from one another by at least 1.3 m on all sides.

Sampling. Effectiveness of the biocontrol treatments was assessed by removing samples of straw from the field plots and estimating the number of pseudothecia of *P. tritici-repentis* per gram of straw. Each plot was sampled by tossing six markers randomly onto plots, then taking straw from a circular area at each marker, as defined by a 57-mm-diameter tulip-bulb planter. Straw from these six areas was bulked to produce one sample per plot for analysis. Straw within a sample was sorted, by visual inspection, into three categories: those with many pseudothecia of *P. tritici-repentis*, those with a moderate number, and those with few or no pseudothecia visible. The straw in each category was weighed, chopped into 1-cm lengths, and mixed thoroughly. A subsample was taken from each category (50 mg or 5% of the category weight, whichever was greater) and was examined with a dissecting microscope at 25× magnification. Pseudothecia of *P. tritici-repentis* in each subsample were counted. Pseudothecia per category of straw were calculated from this information, and the total pseudothecia per gram was calculated by dividing this total by the total sample weight. In 1989 and 1990, the procedure was modified by using two categories (high and moderate/low) instead of three.

Weight of straw per unit area of each plot was also determined in 1988, 1989, and 1990 from the measured weight of the sample and the area sampled.

Plots were sampled during November or December of each year. Previous observations (W. F. Pfender, W. Zhang, and A. Nus, *unpublished data*) indicated that the number of pseudothecia on straw does not increase after November in Kansas.

Data were analyzed by analysis of variance, using the appropriate model (CRD or RCB) for the field design. Means separation was carried out with Duncan's new multiple range test.

Tests of application methods. Although bran inoculum applied as a slurry adhered fairly well to the straw, it was too heavy for practical use. Dried bran adhered well initially to wet straw but later could be removed partially by wind, rain, or irrigation. As a result, we tested the retention of biocontrol fungi in bran

when the colonized bran was applied in combination with alginate to straw. We also tested bran milled to different sizes before application to determine the importance of bran size on retention and survival of biocontrol fungi. As a result, there were four treatments for the factorial combinations of alginate vs. water and large vs. small bran.

Chopped wheat straw (collected from rotary-mowed wheat stubble; described above) was spread onto 1-m^2 plots at approximately 3,000 kg/ha. Inocula of several biocontrol fungi were prepared, as described previously, by growing inocula on wheat bran that was either unmilled or milled to pass through a sieve with 1.0-mm openings. After colonization, the bran was air-dried, then either ground in a blender to pass through the 1.0-mm sieve ("small" bran) or crushed to separate the flakes ("large" bran). The colonized bran was applied to the straw by one of two methods. For alginate treatments, the procedure described previously for inoculation of plots in 1990 was followed. For water treatments, the straw was first sprayed with water at 92 L/ha and then was dusted with the dry-bran preparation. Each plot received 17 g of dry-bran inoculum per square meter. Three fungi were tested, and there were three replicate plots for each fungus/application combination. Treatments were arranged in the field in a CRD. The experiment was performed twice during the summer of 1988.

To assess retention of bran, 10 30-cm-long straws were marked at one end with paint and placed in each plot before application of the inoculum. Immediately after application and at weekly intervals, these straws were rated visually for the amount of bran retained. Ratings were performed by comparing the straws with a set of standards made by dusting known weights of milled bran onto straws; standards were preserved by spraying them with varnish. Four levels comprised the standards with ratings of 0, 1, 3, and 5 corresponding to 0, 1.5, 3, and 6 mg/10-cm straw, respectively.

To determine the survival of the fungi, five nonpainted straws were recovered from each plot 2 wk after inoculation. These straws were returned to the laboratory, where two flakes of bran were removed from each straw and plated onto potato-dextrose agar amended with 100 ppm of chloramphenicol. The petri dishes were incubated at 20 C under alternating 12-h periods of light and dark for 7 days. Plates were examined periodically using a microscope to determine the proportion of bran flakes from which the biocontrol fungus grew.

TABLE 1. Pseudothecial production by *Pyrenophora tritici-repentis* on straw treated with biocontrol fungi in 4 yr of field experiments

	Pseudothecia per gram of straw[z]			
Treatment	1987	1988	1989	1990
Check	1130 a	1149 a	344 a	514 ab
Pithomyces chartarum	NT	1154 a	306 ab	606 a
Limonomyces roseipellis	220 b	459 b	295 ab	169 c
Sterile II	NT	610 b	190 b	273 b
Laetisaria arvalis	NT	NT	200 b	613 a

[z]Values within a column followed by the same letter do not differ ($P = 0.05$) by Duncan's new multiple range test. NT = not tested.

The effect of the application method on survival of fungi and on retention of inoculum at each sampling time was determined by analysis of variance. Means separation was carried out using Duncan's new multiple range test.

RESULTS

The number of pseudothecia per gram of straw in the nontreated plots varied among the 4 yr of the field tests (Table 1). As was expected, application of *P. chartarum* did not significantly decrease pseudothecial production by *P. tritici-repentis*. *L. arvalis* was inconsistent in its suppression of *P. tritici-repentis*. *L. arvalis* was ineffective in 1990, and in 1989, it reduced pseudothecial production below that in the noninoculated check but not below that in the negative check (*P. chartarum*). *L. roseipellis* significantly decreased pseudothecial production by the tan spot pathogen in 3 of the 4 yr of field tests (Table 1). Pseudothecial production on straw treated with this fungus was 20, 40, and 33% of the check production in 1987, 1988, and 1990, respectively. Sterile II significantly reduced pseudothecial production in 2 of the 3 yr. In 1990, when pseudothecia on straw treated with this antagonist were not significantly reduced, three of the four replications of this treatment had very low pseudothecial production, but one replicate showed pseudothecial production equivalent to that in the check.

The amount of residue (gram per square meter) in the plots did not differ significantly among treatments in any year. Values for 1988, 1989, and 1990 were 140, 336, and 388 g/m^2, respectively. From the data in Table 1, inoculum density can be calculated as 161,000 pseudothecia per square meter for the check and 64,000 pseudothecia per square meter for plots inoculated with *L. roseipellis* in 1988. The respective numbers for 1989 were 116,000 and 99,000; those for 1990 were 199,000 and 66,000.

Coverage and retention of the bran-based inoculum on straw in the field was affected by the method of application. At each sampling time, there was no effect of fungus isolate on the degree of inoculum coverage nor any significant fungus-application method interaction. The main effect for application method (Table 2) was significant at each sampling date, and the use of alginate with milled bran provided the greatest coverage. In this experiment, alginate with milled bran retained 73% of its initial coverage after 2 wk; milled bran applied without alginate retained only 37%. Average survival of the biocontrol fungi in the bran was not significantly affected by application method and ranged from 70 to 80%. Survival was, however significantly affected by fungus isolate: *L. arvalis* or *L. roseipellis* was recovered from 94 and 90% of bran pieces, respectively, whereas Sterile II survival was significantly less (44%). When the experiment was repeated, similar results were observed: 1) milled bran with alginate gave superior coverage after 2 wk (although unmilled bran with alginate was not significantly lower), 2) there was no effect of fungus isolate on coverage, and 3) Sterile II showed significantly lower survival in bran than did the other two fungi.

DISCUSSION

The research reported here demonstrates that it is possible to reduce the primary inoculum of a phytopathogen in surface-borne

TABLE 2. Comparison of methods of application of biocontrol fungus inoculum to straw in the field

Formulation[x]		Coverage[y]			Retention (%)	Survival[z]
Bran size	Liquid	0 wk	1 wk	2 wk	(2 wk)	(%)
S	Alginate	4.4 a	4.4 a	3.2 a	73	71 a
S	Water	3.5 a	2.6 b	1.3 b	37	80 a
L	Alginate	1.5 b	1.7 bc	0.7 b	47	82 a
L	Water	1.1 b	0.7 c	0.2 b	18	71 a

[x]Colonized bran was milled to small size (S) or left unmilled (L) and was dusted onto straw wetted with water or alginate.
[y]Coverage rated visually on a 0–5 scale: 0 = no bran visible, 5 = approximately 0.6 mg of bran per 1 cm of straw length (discussed in text). Ten straws per replicate, three replicates per treatment per fungus. Values are averages for the three fungi, and there were no fungus or fungus × application method effects. Within a column, values followed by the same letter do no differ ($P = 0.05$) by Duncan's new multiple range test.
[z]The percentage of bran pieces from which the applied biocontrol fungus could be recovered 2 wk after application.

residue using a biological control agent applied to crop residue in the field. The success of this approach in reducing yield loss will be affected by a number of factors related to the epidemiological characteristics of the target disease, to environmental conditions after application, and to formulation and application of the biocontrol agent.

For tan spot of wheat, epidemic severity and yield loss are responsive to primary inoculum level (1). However, the data show that reductions of less than an order of magnitude in primary inoculum are not likely to effect disease control. Yield loss was reduced to background level when inoculum was decreased from 25,000 to 2,500 pseudothecia per square meter or from 5,000 to 500 pseudothecia per square meter in two different years (1). In the research reported here, inoculum density of *P. tritici-repentis* was extremely high (because of intentional manipulation of disease severity on the crop before harvest), and application of *L. roseipellis* reduced primary inoculum density (pseudothecia per square meter) only by a factor of 2.5–3.0, from 161,000 to 64,000 in 1988 and from 199,000 to 66,000 in 1990. Although this reduction might alleviate yield loss to some extent, it would not control the disease to levels expected with complete elimination of the local primary inoculum. For polycyclic diseases, such as tan spot, a more complete reduction in residue-borne primary inoculum is required for effective control. Monocyclic diseases, such as Cephalosporium stripe of wheat, may be better candidates for biocontrol of surface residue-borne inoculum, because inoculum reductions will be more closely reflected in disease decreases.

For effective biocontrol of a residue-borne pathogen, such as *P. tritici-repentis*, that produces primary inoculum from the mycelium already present in infested straw at harvest, application of the treatment must be timely and thorough. We found in related field experiments (W. F. Pfender, W. Zhang, and A. Nus, *unpublished data*) that application of *L. roseipellis* to straw at progressively later times after wheat harvest produced progressively poorer reductions in *P. tritici-repentis* pseudothecia, such that biocontrol was ineffective when inoculum was applied more than 4 wk after harvest. In addition, because the antagonist must act quickly to suppress the pathogen, success depends on thorough coverage of the straw so the antagonist can quickly reach most sites of pathogen infestation. This can be achieved by applying a large amount of inoculum per field area or by developing a suitable method for evenly distributing a smaller amount of finely divided inoculum. We used a dry, finely ground preparation of bran-based inoculum to get fairly even, though incomplete, coverage of straw (220 kg/ha). Other methods could be developed to achieve better coverage with an equal or lower amount of inoculum. For example, a device for treatment of the straw as it leaves the combine might provide more thorough coverage of straw.

Another critical aspect of this biocontrol approach is retention of viable biocontrol inoculum on the surface-borne residue through weather extremes: heavy rain, wind, and high temperatures. To retain inoculum long enough for the biocontrol agent to effectively colonize the straw, we used alginate. In replicated but unrepeated observations, we found that the extent of straw colonization by the biocontrol agent (measured as the percentage of sampled straw fragments) was higher with the alginate/milled bran formulation than with our other formulations. Other methods could be developed to achieve better retention. Another factor that influences retention is the growth rate and mycelial character of the antagonistic fungus. Fungi capable of rapid growth, particularly those capable of responding quickly to transient episodes of adequate moisture, will anchor themselves more certainly than slower growing fungi. One aspect of formulation and application we did not explore is the use of materials to increase moisture content or wetness duration of the straw. We previously demonstrated (7) that prolonged periods of high moisture favor antagonism to *P. tritici-repentis* in straw. If the biocontrol agent was applied in (or followed by) a material that retains moisture, colonization of the straw and subsequent antagonism would be improved. Related field trials (W. F. Pfender, W. Zhang, and A. Nus, *unpublished data*) provided no evidence for improvement of colonization or biocontrol by *L. roseipellis* when applied with invert emulsions (10), but other approaches might be successful.

Biocontrol is most likely to be successful if the microbial community is manipulated to enhance the antagonistic interactions that occur naturally in the target system. Therefore, mechanisms of antagonism are of interest in developing biocontrol. The mechanism of pathogen inhibition by the fungi we applied is not known with certainty, but in previous laboratory work, we demonstrated that *L. roseipellis* is chitinolytic (9) and can invade and destroy the mycelium of *P. tritici-repentis* (5). Such mycoparasitism is congruent with the ecological characteristics of crop residue (the lack of primary production and the consequent reduction in nutrient availability with time), because nitrogen (a limiting nutrient) would be available in the chitin. Another type of interaction that might be exploited for biocontrol is the production of antibiotics toxic to the phytopathogen. We have no evidence that any of the fungi we tested produce antibiotics toxic to *P. tritici-repentis*. However, fungi or bacteria capable of such activity might be found. Because antibiotic production often occurs during secondary metabolism on adequate nutrient sources, crop residue may not provide a substrate favorable for antibiotic production. However, the required nutrient base could be supplied as part of the formulation of antibiotic-producing antagonists. A third possible approach is competition for nutrients between the residue-borne pathogen and an applied biocontrol agent. In other research, we found that pseudothecial production by *P. tritici-repentis* in a cellulose-based culture medium was proportional to nitrogen content (8). Depending on the timing of nitrogen intake from straw by *P. tritici-repentis* and its exposure to nutrient competition after wheat harvest, it might be possible to exploit nitrogen competition as a means of suppressing primary inoculum production.

If successful, biocontrol of phytopathogens in surface-borne crop residues will be useful, particularly in conservation-tillage crop production, in which residue-borne pathogens present a major constraint for some crops. Our results indicate that biocontrol may be effective for residue-borne *P. tritici-repentis* and suggest that it may be effective for other pathogens as well, particularly those that cause monocyclic diseases and/or diseases for which some amount of disease is acceptable. For biocontrol of residue-borne pathogens to provide adequate yield protection, however, advances are needed in formulation and application methods. The challenge is to obtain adequate coverage of crop residue with a small amount of inoculum, to improve retention, and to enhance the moisture-retention capability of the residue. More effective antagonistic fungi or bacteria may also be discovered, selected, or generated. Litter-inhabiting microbes, such as *L. roseipellis*, are likely to be best adapted for activity under the physical and ecological constraints of surface-borne crop residue. *L. roseipellis* was selected from such a community, and it has performed moderately well. A fungus with better tolerance to moisture stress, however, would be more effective in this environment.

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