

# EVALUATION OF A BIOLOGICAL AGENT FOR CONTROLLING BASIDIOMYCETE ATTACK OF DOUGLAS-FIR AND SOUTHERN PINE<sup>1</sup>

*Jeffrey J. Morrell and Camille M. Sexton*

Assistant Professor and Research Assistant  
Department of Forest Products, College of Forestry  
Oregon State University, Corvallis, OR 97331

(Received October 1988)

## ABSTRACT

A biological control agent (Binab®) containing *Trichoderma polysporum* and *T. harzianum* was evaluated for its ability to prevent or arrest attack of loblolly pine (*Pinus taeda*) sapwood and Douglas-fir (*Pseudotsuga menziesii*) heartwood by five Basidiomycetes commonly isolated from poles in service. Studies of *Lentinus lepideus* were included for comparison. In general, the biocontrol agents performed well against *L. lepideus* and other brown-rot fungi out of ground contact, but they did not completely eliminate most of the test fungi. The biological control agents had little effect on *L. lepideus* when the wood was exposed to soil. In addition, the biocontrol agents had little effect on white-rot fungi, which are an important component of the microflora in decaying poles. Results suggest that Binab® is not suitable for remedial decay control without supplemental treatments that favor growth and activity of *Trichoderma*.

**Keywords:** Biological control, wood decay, southern pine, Douglas-fir, Basidiomycetes, *Trichoderma harzianum*, *Trichoderma polysporum*.

## INTRODUCTION

Although toxic chemicals remain the primary method used to limit the degradation of wood in adverse environments, increased environmental regulation and a desire for decreased chemical dependence have fostered renewed interest in the use of biological agents to control decay fungi. Biological control agents have been successfully applied to some agricultural systems (Baker and Cook 1974; Papavizas 1981, 1985; Baker 1987); however, the need for long-term control of decay, coupled with intolerance of any failure of the biological control agent, has limited the use of biocontrol agents in wood products.

The potential effectiveness of biological agents in this role was initially reported by Ricard and Bollen (1967), who used *Scytalidium* Strain FY to prevent colonization of Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco) by *Poria carbonica* Overholts and *Postia placenta* (Fr.) M. Lars et Lomb. Field studies indicated that protection was incomplete, and the treatment was never commercialized in the United States (Graham 1973). Biocontrol agents have also been advocated for preventing colonization of freshly cut stumps (Kallio 1971, 1972) and logs in storage (Hulme and Shields 1972). None of these agents, however, appear to be widely used.

The recently developed biocontrol formulation Binab®, which contains *Trichoderma polysporum* (Link) Rifai and *T. harzianum* Rifai (Ricard 1976; Morris

---

<sup>1</sup> This is Paper 2392 of the Forest Research Laboratory, Oregon State University, Corvallis. Mention of trade names does not constitute endorsement by Oregon State University.

et al. 1984), has received extensive attention in Europe. Although results have been conflicting, several tests indicate that the biological agent can control *Len-tinus lepideus*, one of the more common decay fungi colonizing creosoted Scots pine (*Pinus sylvestris* L.). Several British utilities have treated test poles with this formulation, and a number of experiments are underway (Morris et al. 1984; Bruce and King 1986a, b).

The *Trichoderma*-based formulation has been shown to strongly inhibit the growth of the common brown-rot fungus *L. lepideus* under controlled laboratory conditions, either because of the production of inhibitory volatiles (Bruce et al. 1984) or the deposition of nonvolatile components in the wood cell wall (Bruce and King 1983). In field tests, the biological agent has produced more variable results (Morris et al. 1984; Bruce and King 1986a, b); however, the organisms are difficult to isolate consistently from wood in the field. In general, *Trichoderma* spp. have not completely colonized the wood substrate to eliminate established decay fungi.

Although the results of studies in Europe have been less than promising, considerable interest remains in the use of biological agents to control decay fungi (Preston et al. 1982). However, the variety of wood species used in the United States and the array of fungi capable of colonizing these different substrates indicate that biocontrol strategies should be implemented cautiously. Douglas-fir and southern pine comprise the most commonly used species for utility poles in the United States. Previous studies suggest that these species are colonized by a variety of white-, brown-, and soft-rot fungi (Eslyn 1970; Graham and Corden 1980; Zabel et al. 1980, 1982). As a result, a potential biocontrol agent must be effective against a broad spectrum of decay agents. Without this effectiveness, less common decay fungi with some tolerance to the biocontrol agent could increase in abundance and relative importance. Before biocontrol agents can be effectively employed, more information is needed on how well these agents limit colonization of commercially important American wood species by common decay fungi. This report details the ability of the *Trichoderma*-based biological control agent Binab<sup>®</sup> to limit growth of 10 Basidiomycetes commonly isolated from Douglas-fir or yellow pine.

#### MATERIALS AND METHODS

The biocontrol formulation Binab<sup>®</sup> AB (Mycotek) was obtained through Agrotek, Inc. (Westport, CT). The pellets, initially presumed to be a semipure mixture of *Trichoderma harzianum* and *T. polysporum*, were subsequently found to contain extensive surface *Mucor* species and bacterial contamination. This contamination tended to inhibit the growth of both *Trichoderma* species but could be reduced by 24-h exposure of whole pellets to ultraviolet light. The pellets were then ground with a mortar and pestle prior to use.

#### *In-vitro* petri plate tests

The ability of the biological agent to inhibit Basidiomycete growth was first tested by inoculating petri dishes containing 20 ml of 1% malt extract or a salt agar (Nilsson 1973) with combinations of the biological agent and one of 10 Basidiomycetes. A 1% malt extract/1% sawdust agar was used for selected tests, but no significant difference was found for growth of fungi on this media and the

TABLE 1. *Basidiomycetes tested against Binab® AB in agar or wood blocks.*

Fungus	Isolate number <sup>a</sup>	Test conditions		Type of fungus <sup>b</sup>
		Agar	Wood	
<i>Bjerkandera adusta</i>	FP-135160-SP (Mad.)	X		W
<i>Coriolus versicolor</i>	R-105 (Mad.)	X	X	W
<i>Crustoderma dryinum</i>	51C Arl. 5-17-82 (OSU)	X		B
<i>Gloeophyllum saepiarium</i>	54UT-18 (OSU)	X		B
<i>Irpex lacteus</i>	FP-105915-SP (Mad.)	X	X	W
<i>Lentinus lepideus</i>	44C (Forintek)	X	X	B
<i>Phlebia radiata</i>	L-15608-SP (Mad.)	X		W
<i>Poria carbonica</i>	1978 (OSU)	X	X	B
<i>Poria xantha</i>	FP-105494-SP (Mad.)	X		B
<i>Postia placenta</i>	FP-94267A (Mad.)	X	X	B

<sup>a</sup> Isolates were obtained from U.S. Forest Products Laboratory in Madison, Wisconsin (Mad.), Oregon State University (OSU), or Forintek Canada Corp., Ottawa, Ontario (Forintek).

<sup>b</sup> W = white-rot fungus, B = brown-rot fungus.

treatment was discontinued. The biological agent was added to a well cut in the center of the agar in each dish. The decay fungi were added as 3-mm-diameter agar disks cut from the edges of actively growing cultures of the test fungi (Table 1). Three disks, equidistantly spaced around the edge, were placed on each plate.

The plates were inoculated in one of three sequences: (A) Biocontrol agent and Basidiomycetes simultaneously; (B) Biocontrol agent first, then Basidiomycetes when the biological agent had grown 5–10 mm; or (C) Basidiomycetes first, then biocontrol agent when the Basidiomycetes had grown 5–10 mm. A total of eight plates of each medium were tested per fungus for each inoculation sequence. Inoculated plates were incubated at 15 or 28 C. The plates were observed, and the radial growth rates of both the biological agent and the Basidiomycete were measured twice before the fungi touched or stopped growing. At the end of the incubation period, the original inoculum plug of each Basidiomycete that failed to grow was subcultured onto fresh 1% malt extract agar to determine whether exposure to the biological agent was lethal.

#### Wood block tests

Tests were made with wood to provide a measure of field performance. Blocks (2.5 cm × 2.5 cm × 10 cm) of Douglas-fir heartwood and of sapwood from loblolly pine (*Pinus taeda* L.), a southern pine, were sterilized (30 min at 121 C), covered on the transverse faces, and waxed. The transverse faces were exposed, and the blocks were soaked in sterile distilled water until they reached 40–50% moisture content. Agar squares (2.5 cm × 2.5 cm) cut from actively growing cultures of *Coriolus versicolor* (L.:Fr.) Quel, *Irpex lacteus* Fr., *Lentinus lepideus*, *Poria carbonica*, or *Postia placenta* were placed on both exposed transverse faces. These squares were held in place by small feeder blocks (2.5 cm × 2.5 cm × 1.25 cm) of the same species, and the entire assemblage was held in place by a rubber band. The procedures closely paralleled those used to test fumigant performance (Morrell and Corden 1988). Ten milligrams of the biological agent were placed in a hole 0.9 cm in diameter and 2.0 cm deep drilled at the center point of each block. The hole was then plugged with a tight-fitting rubber septum.

The blocks were inoculated according to the same three sequences (A, B, and

C) as were the petri plates, except that a 2-week period elapsed between the addition of the first and second agents. In addition, control blocks containing only the biological agent or the particular decay fungus were prepared. Each Basidiomycete/biocontrol agent combination was replicated on three loblolly pine and six Douglas-fir blocks. These blocks were compared to similar blocks inoculated with only the biological agent or the Basidiomycete. The inoculated blocks were then incubated at 15 or 28 C for 4 weeks in sealed chambers. An additional three loblolly pine and three Douglas-fir blocks were inoculated with the same fungi and incubated for 8 weeks at each temperature.

Effectiveness of the biological agent was assessed by removing the feeder blocks and cutting a series of 0.3-cm-thick sections from each end of the larger block. The outer section from each end was discarded and the next two sections were retained. In addition, two sections were cut from the zone adjacent to each side of the biological treatment hole. These sections were cut into a series of 16 cubes (0.6 cm). The inner four pieces of each section were plated onto media containing 2.5% malt extract and 1.0% agar (MEA) or MEA amended with 10 ppm Benomyl to retard growth of the *Trichoderma* species. The plates were observed for at least 28 days for evidence of fungal growth, which was examined for characteristics typical of the test fungus.

#### *Soil block tests*

Small block tests provide a relative measure of fungal survival out of direct ground contact; however, they do not allow for accurate determination of the effects of fungal interactions on wood properties such as weight. Following procedures outlined by the American Society for Testing and Materials (1986), we used soil block tests to determine the effects of the biocontrol agent on weight losses caused by *C. versicolor*, *I. lacteus*, *L. lepideus*, *Postia placenta*, or *Poria carbonica*. French square bottles (454 ml) were filled with 165 g of soil (60% moisture content), and a western hemlock (*Tsuga heterophylla* [Raf.] Sarg.) feeder was placed on the wood surface. The bottles were then capped and autoclaved (121 C) for 45 min, cooled overnight, and reautoclaved (121 C) for 15 min to eliminate spore-forming bacteria. After cooling, the chambers were inoculated with the biological control agent or one of the five Basidiomycetes in the following sequences: (A) Basidiomycete and biocontrol agent simultaneously; (B) biocontrol agent first, Basidiomycete 2 weeks later; and (C) Basidiomycete first, biocontrol agent 2 weeks later. In addition, the Basidiomycete alone and the biological control agent alone were used as control inoculations.

The bottles were incubated at 28 C for 2 weeks to allow the first fungus to become established. In tests with multiple inoculations, the second fungus was incubated for 7 days. Loblolly pine sapwood and Douglas-fir heartwood test blocks (1.90 cm<sup>3</sup>) were then aseptically placed, two per bottle, on the feeder. The blocks were oven-dried (54 C for 24 h), weighed (0.001 g), and steamed for 20 min at 100 C prior to placement in the bottles. The blocks were incubated at 28 C for 12 weeks. At the conclusion of the study, the blocks were gently scraped clean of adhering soil and mycelium and oven-dried for 24 h (54 C) prior to weighing. Lack of weight loss over the fungus exposure period was used as a measure of effectiveness of the biological control agent.

TABLE 2. Average radial growth of the biocontrol agent (BCA) and 10 Basidiomycetes in inoculation sequences A, B, and C on 1% malt extract and Nilsson's salt agar incubated at 15 and 28 C.<sup>a</sup>

Fungus <sup>b</sup>	Average radial growth (% of control) <sup>c</sup>											
	Inoculation sequence A				Inoculation sequence B				Inoculation sequence C			
	15 C		28 C		15 C		28 C		15 C		28 C	
	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus
	1% Malt extract agar											
<i>B. adusta</i>	80	38	0	51	93	103	88	15	0	115	68	22
<i>C. versicolor</i>	42	13	22	38	88	5 (50)	77	33	0	156	0	88
<i>C. dryinum</i>	11	165	37	20	( <sup>d</sup> )	( <sup>d</sup> )	84	17	( <sup>d</sup> )	( <sup>d</sup> )	5	32
<i>G. saepiarium</i>	6	78	12	56	71	2 (50)	81	9	0	74	11	46
<i>I. lacteus</i>	8	18	12	16	65	105	92	30	0	74	0	54
<i>L. lepideus</i>	119	81	31	24	52	0 (40)	67	36	8	59	21	38
<i>P. radiata</i>	16	44	12	19	60	133	87	47	0	184	6	89
<i>P. carbonica</i>	97	2	56	46	81	12	68	10	11	24	24	96
<i>P. placenta</i>	47	18	43	44	56	66	44	17	18	104	13	39
<i>P. xantha</i>	37	11	26	28	39	80	103	4	3	355	2	35
	Nilsson's salt agar											
<i>B. adusta</i>	2	64	7	17	31	0 (50)	79	0 (100)	12	47	( <sup>d</sup> )	( <sup>d</sup> )
<i>C. versicolor</i>	0	176	54	19	68	0 (0)	77	18	16	74	34	33
<i>C. dryinum</i>	0	136	54	0 (100)	( <sup>d</sup> )	( <sup>d</sup> )	97	0 (50)	0	136	( <sup>d</sup> )	( <sup>d</sup> )
<i>G. saepiarium</i>	36	71	51	26	84	0 (0)	71	0 (10)	80	31	47	44
<i>I. lacteus</i>	0	59	10	12	102	0 (50)	108	0 (10)	14	16	20	37
<i>L. lepideus</i>	19	53	37	3	84	0 (0)	73	9	31	72	44	36

TABLE 2. Continued.<sup>a</sup>

Fungus <sup>b</sup>	Average radial growth (% of control) <sup>c</sup>											
	Inoculation sequence A				Inoculation sequence B				Inoculation sequence C			
	15 C		28 C		15 C		28 C		15 C		28 C	
	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus	BCA	Fungus
<i>P. radiata</i>	25	31	29	12	70	0 (70)	94	0 (60)	13	24	94	22
<i>P. carbonica</i> <sup>e</sup>	123	171	—	—	( <sup>d</sup> )	( <sup>d</sup> )	—	—	61	79	—	—
<i>P. placenta</i>	32	31	24	61	96	0 (0)	90	0 (10)	25	99	15	591
<i>P. xantha</i> <sup>e</sup>	31	24	—	—	( <sup>d</sup> )	( <sup>d</sup> )	—	—	43	224	—	—

<sup>a</sup> Inoculation sequences were (A) biocontrol and Basidiomycete simultaneously, (B) biocontrol first, followed by Basidiomycete, and (C) Basidiomycete followed by the biocontrol.

<sup>b</sup> See Table 1. *P.* refers to *Poria* except in the case of *Postia placenta*.

<sup>c</sup> Control is based upon percentage of growth of the test fungus in the presence of Binab® in comparison with growth in the absence of the biocontrol agent. Numbers in parentheses represent percentage of survival of fungus as determined by subculturing.

<sup>d</sup> Not tested.

<sup>e</sup> *Poria carbonica* and *Poria xantha* failed to grow on Nilsson's media at 28 C.

## RESULTS AND DISCUSSION

*Petri plate tests*

Growth measurements in petri dishes containing both the biocontrol agent and test fungi indicated that the biocontrol agent had limited effect on the growth rate and survival of Basidiomycetes (Table 2). In malt extract agar, only *L. lepideus*, *Gloeophyllum saepiarium* (Wulf.:Fr.) Karst., and *C. versicolor* were completely inhibited (growth rate less than 5% of control) in inoculation sequence B (biocontrol agent first, then Basidiomycetes). Only *Bjerkandera adusta* (Willd.:Fr.) Karst. was not inhibited by prior addition of the biological agent. None of the test fungi were completely inhibited in inoculation sequence A (biocontrol agent and Basidiomycetes simultaneously). Conversely, the growth rate of the *Trichoderma* species declined substantially in inoculation sequence C (Basidiomycetes first, then biocontrol agent). The inability to grow in the presence of an actively growing Basidiomycete suggests that the biological agent will be unable to arrest actively growing decay pockets in wood; however, malt extract contains an abundant sugar source that may alter fungal growth rates. In wood, these free sugars would not be available to the Basidiomycete, and this unavailability might affect its growth rate and interaction with the control agent. Although growth of some fungi on malt extract agar was inhibited by the biological agent, none of the fungi were killed by exposure to *Trichoderma* species. This lack of mortality indicates that the ability of the biological agent to completely control decay fungi in vitro is limited.

The effectiveness of the biocontrol agent on Basidiomycete growth was limited on malt extract but was more substantial on the salt agar. This effect was particularly noticeable in inoculation sequence B. In addition, exposure to the biological agent was lethal to nearly all of the test fungi when the plates were incubated at 15 C. Although fungal control was less dramatic in inoculation sequences A and C, growth rates on Nilsson's salt agar were substantially lower than those on MEA.

The results of tests on salt agar appear promising; however, Basidiomycete control was not complete in inoculation sequences A or C. These results suggest that the biological agent would not be effective as a remedial decay control agent but might provide protection against fungal invasion of sound wood uncolonized by Basidiomycetes.

*Wood block tests*

The small block tests indicated that none of the four test fungi were completely inhibited by the biological agent, even after a 2-month incubation period (Tables 3, 4). In addition, *Trichoderma* species failed to uniformly colonize blocks of either wood species. Cultures from some blocks were characterized by the presence of microfungi, notably *Penicillium* species, that appeared to inhibit *Trichoderma* species and to have minimal effect on growth of the Basidiomycetes. These contaminants appeared to be associated with the pellets and could limit field effectiveness.

After a 1-month incubation period, *Trichoderma* species were isolated from 70% of the cubes removed from all of the loblolly pine test blocks; 19% of these same cubes revealed the presence of viable Basidiomycetes (Table 3). After a

TABLE 3. Fungus survival in southern pine blocks inoculated in sequences A, B, and C with the biocontrol agent in combination with selected Basidiomycetes and incubated for 1 or 2 months at 15 and 28 C.<sup>a</sup>

Fungus <sup>b</sup>	Percent survival (Basidiomycete/biocontrol agent) <sup>c</sup>					
	Inoculation sequence A		Inoculation sequence B		Inoculation sequence C	
	15 C	28 C	15 C	28 C	15 C	28 C
	One month					
<i>C. versicolor</i>	52 (58)	0 (96)	8 (100)	30 (96)	0 (100)	50 (0)
<i>I. lacteus</i>	75 (36)	26 (43)	17 (100)	0 (100)	0 (100)	43 (14)
<i>L. lepideus</i>	28 (50)	0 (100)	6 (85)	0 (98)	0 (100)	43 (0)
<i>P. carbonica</i>	50 (88)	5 (100)	19 (74)	0 (52)	0 (100)	15 (23)
<i>P. placenta</i>	14 (21)	0 (100)	0 (100)	0 (40)	23 (96)	60 (0)
Mean	46 (51)	8 (89)	10 (95)	4 (78)	2 (99)	43 (7)
	Two months					
<i>C. versicolor</i>	0 (95)	0 (89)	10 (94)	12 (76)	0 (100)	62 (0)
<i>I. lacteus</i>	9 (100)	65 (26)	26 (100)	34 (36)	0 (100)	84 (0)
<i>L. lepideus</i>	2 (98)	0 (100)	0 (100)	0 (54)	0 (100)	94 (8)
<i>P. carbonica</i>	30 (88)	0 (84)	0 (96)	0 (44)	0 (100)	0 (2)
<i>P. placenta</i>	0 (88)	0 (96)	0 (100)	0 (96)	0 (100)	0 (0)
Mean	7 (95)	18 (79)	8 (99)	12 (61)	0 (100)	63 (2)

<sup>a</sup> Inoculation sequences were (A) biocontrol and Basidiomycete simultaneously, (B) biocontrol first, followed by Basidiomycete, and (C) Basidiomycete followed by the biocontrol.

<sup>b</sup> See Table 1. *P.* refers to *Poria* except in the case of *Postia placenta*.

<sup>c</sup> As a percentage of isolations from blocks inoculated with only one of the test fungi. Values represent survival of Basidiomycete; figures in parentheses represent percentage of survival of the biocontrol agent (Binab®). Each treatment was replicated on 3 blocks.

2-month incubation, 18% of the cubes contained decay fungi, and 73% contained *Trichoderma* species. These results suggest that little active colonization occurred after the initial invasion and indicate that the biological control agent was unable to completely control the decay fungi. This effect was most noticeable in inoculation sequences A and C. *Lentinus lepideus* and *P. placenta* were both inhibited by the biological agent under most of the test conditions; however, even these fungi survived under certain conditions. Of the five test fungi, *I. lacteus* appeared to be least affected by the biological agent. This white-rot fungus is commonly isolated from southern yellow pine utility poles (Zabel et al. 1980, 1982). Although studies have suggested that white-rot fungi are less sensitive to Binab® (T. L. Highley unpublished data) than are other Basidiomycetes, *C. versicolor*, a white-rot fungus, proved to be more sensitive. These differences suggest subtle variations in physiology that warrant further investigation. *Poria carbonica* exhibited some tolerance to the biological agent after a 1-month exposure period but appeared to succumb after an additional month of incubation. This fungus is a common inhabitant of Douglas-fir utility poles (Esllyn 1970; Graham and Corden 1980), and its ability to survive exposure to the biological agent, even at low levels, should be of concern.

Although the biological agent colonized Douglas-fir at lower levels than it did southern pine, the degree of Basidiomycete inhibition appeared to be similar in both species (Table 4). In addition, the sequence of inoculation, which had no consistent effect on survival in the southern pine, appeared to influence Basidiomycete survival in Douglas-fir. Blocks inoculated under sequence C had higher levels of colonization by fungi at both sampling points than blocks inoculated under either sequence A or sequence B. This pattern suggests that the biological

TABLE 4. Fungus survival in Douglas-fir blocks inoculated with the biocontrol agent in sequences A, B, and C in combination with selected Basidiomycetes and incubated for 1 or 2 months at 15 and 28 C.<sup>a</sup>

Fungus <sup>b</sup>	Percent survival (Basidiomycete/biocontrol agent) <sup>c</sup>					
	Inoculation sequence A		Inoculation sequence B		Inoculation sequence C	
	15 C	28 C	15 C	28 C	15 C	28 C
One month						
<i>C. versicolor</i>	5 (95)	0 (5)	1 (45)	11 (93)	4 (63)	85 (20)
<i>I. lacteus</i>	15 (51)	0 (32)	0 (40)	82 (100)	22 (42)	100 (0)
<i>L. lepideus</i>	0 (54)	6 (100)	0 (47)	43 (61)	43 (40)	46 (93)
<i>P. carbonica</i>	62 (99)	2 (7)	33 (59)	21 (100)	18 (65)	9 (81)
<i>P. placenta</i>	22 (100)	14 (16)	9 (46)	46 (73)	26 (25)	20 (27)
Mean	20 (81)	6 (36)	8 (47)	41 (85)	21 (47)	52 (51)
Two months						
<i>C. versicolor</i>	0 (100)	0 (59)	0 (81)	5 (100)	15 (27)	— (—) <sup>d</sup>
<i>I. lacteus</i>	19 (46)	0 (80)	0 (42)	27 (72)	19 (49)	100 (0)
<i>L. lepideus</i>	0 (68)	0 (87)	0 (25)	0 (100)	48 (73)	100 (83)
<i>P. carbonica</i>	23 (100)	14 (28)	58 (44)	0 (100)	14 (81)	41 (10)
<i>P. placenta</i>	0 (100)	18 (56)	52 (83)	43 (83)	0 (26)	61 (40)
Mean	8 (86)	6 (64)	21 (55)	15 (91)	18 (52)	76 (33)

<sup>a</sup> Inoculation sequences were (A) biocontrol and Basidiomycete simultaneously, (B) biocontrol first, followed by Basidiomycete, and (C) Basidiomycete followed by the biocontrol.

<sup>b</sup> See Table 1. *P.* refers to *Poria* except in the case of *Postia placenta*.

<sup>c</sup> As a percentage of isolations from blocks inoculated with only one of the test fungi. Values represent survival of Basidiomycete; figures in parentheses represent percent survival of the biocontrol agent (Binab<sup>®</sup>). Each treatment was replicated on three blocks.

<sup>d</sup> Not tested.

agent is unable to overcome established decay fungi and might be ineffective in arresting existing decay pockets. *Irpex lacteus* also exhibited considerable resistance to the biological agent in Douglas-fir blocks, while *L. lepideus* was only able to survive at high levels (>20%) when it was inoculated prior to the *Trichoderma* species. In spite of the lower levels of survival following a 2-month incubation period, none of the Basidiomycetes were completely controlled by application of the biological agent. Effective decay control of high-value wood products requires complete Basidiomycete control; therefore, this biocontrol agent is not likely to be generally accepted by users of either wood species unless supplemental treatments that enhance its performance are identified.

#### Soil block tests

As expected, weight losses were generally higher on southern pine sapwood than on the moderately durable Douglas-fir heartwood. The soil block tests indicated that the biocontrol agent had a substantial impact on the weight losses in both Douglas-fir and loblolly pine blocks (Table 5). Even *I. lacteus*, which appeared resistant in the small block tests, produced lower weight losses when the biocontrol agent was present. This effect was most noticeable in inoculation sequence A. Similar results were obtained with *C. versicolor*, the other white-rot fungus tested. In inoculation sequence C, the biocontrol agent appeared to enhance weight loss from *L. lepideus* and *P. placenta* on southern pine. The nature of this effect remains unclear.

Of the three brown-rot fungi, *L. lepideus* appeared to be least affected by the

TABLE 5. Average weight losses (SD) of Douglas-fir heartwood and loblolly pine sapwood blocks exposed to selected Basidiomycetes and a biological control agent in inoculation sequences A, B, and C.<sup>a</sup>

Fungus <sup>b</sup>	Wood weight loss (%)			
	Inoculation sequence A	Inoculation sequence B	Inoculation sequence C	Control
	Loblolly pine			
<i>C. versicolor</i>	2.63 (0.19)	3.32 (0.24)	14.00 (4.30)	18.74 (5.41)
<i>I. lacteus</i>	5.28 (2.09)	14.75 (6.77)	21.58 (4.30)	16.20 (4.06)
<i>L. lepideus</i>	34.13 (13.66)	36.78 (2.90)	56.11 (7.63)	40.80 (6.06)
<i>P. carbonica</i>	2.52 (0.27)	3.24 (0.28)	2.96 (0.22)	17.21 (8.01)
<i>P. placenta</i>	2.73 (0.40)	3.32 (0.13)	59.98 (3.86)	42.76 (15.13)
				3.75 (0.30) <sup>c</sup>
	Douglas-fir			
<i>C. versicolor</i>	2.11 (0.22)	3.05 (0.27)	2.13 (0.24)	2.47 (0.25)
<i>I. lacteus</i>	2.00 (0.26)	3.06 (0.24)	2.82 (0.26)	3.34 (1.99)
<i>L. lepideus</i>	2.36 (0.38)	5.07 (6.20)	20.43 (9.16)	30.37 (5.06)
<i>P. carbonica</i>	1.75 (0.62)	2.40 (0.46)	8.44 (10.63)	7.54 (3.81)
<i>P. placenta</i>	2.02 (0.32)	2.84 (0.49)	15.57 (3.50)	32.69 (6.87)
				2.35 (0.32) <sup>c</sup>

<sup>a</sup> Values represent means of six replicates. Inoculation sequences were (A) biocontrol and Basidiomycete simultaneously, (B) biocontrol first, followed by Basidiomycete, and (C) Basidiomycete followed by the biocontrol.

<sup>b</sup> See Table 1. *P.* refers to *Poria* except in the case of *Postia placenta*.

<sup>c</sup> Soil block tests with Binab<sup>®</sup> alone.

biocontrol agent. This lack of effect was especially curious because the biocontrol agent performed well against this fungus in the small block and petri plate tests. In addition, it has successfully controlled *L. lepideus* in the field (Bruce et al. 1984). Apparently, the high moisture levels or the direct soil contact limited the effectiveness of the control agent. This failure may help explain the poor field performance by this agent.

The weight losses indicate that the presence of the biocontrol agent reduces decay by *C. versicolor*, *Poria carbonica*, and *Postia placenta* on both southern pine and Douglas-fir. These results, coupled with the survival of Basidiomycetes exposed to the biocontrol agents in the small block tests, suggest that the *Trichoderma* species tested may have subtle effects on Basidiomycete physiology.

#### CONCLUSIONS

The results indicate that Binab<sup>®</sup> can inhibit growth of selected Basidiomycetes on malt extract agar and the salt media; however, this treatment did not completely inhibit all of the test fungi. Similarly, the biocontrol agent colonized Douglas-fir and southern pine blocks; however, it neither completely inhibited the growth of the five selected Basidiomycetes nor uniformly colonized the wood. Uniform colonization is essential for long-term wood protection and should be considered a high-priority characteristic of any biological control agent. Of the fungi tested, *L. lepideus* was controlled to the greatest degree in nonsoil contact. This species has been the subject of extensive study in Europe where biocontrol agents have been used commercially, but application of a biological agent to control decay of preservative-treated southern pine and Douglas-fir appears limited. Recent studies suggest that a variety of white- and brown-rot fungi play a prominent role in the decay of Douglas-fir and southern pine poles (Esllyn 1970; Graham and Corden

1980; Zabel et al. 1980, 1982). As a result, potential biocontrol agents must be effective against a range of fungi. Our results suggest that Binab<sup>®</sup> has less widespread effectiveness than previously reported, particularly against white-rot fungi. Furthermore, environmental conditions have a substantial effect on the performance of the biological agent. Thus, Binab<sup>®</sup> does not appear to be a feasible method for preventing or controlling decay of Douglas-fir or southern pine poles unless supplemental treatments are identified to inhibit other fungi and stimulate colonization by the control agent.

#### ACKNOWLEDGMENTS

The authors wish to acknowledge Agrotek, Inc., Westport, CT for supplying material and partial financial support for this research. Portions of this study were completed under USDA Competitive Research Grant 87 FSTY-9-0276.

#### REFERENCES

- AMERICAN SOCIETY FOR TESTING AND MATERIALS. 1986. Standard method of testing wood preservatives by laboratory soil block culture. D1413-76. *In* Annual book of ASTM standards, Sect. 4, Vol. 04.09 Wood. Philadelphia, PA.
- BAKER, K. F. 1987. Evolving conceptual biological control of plant pathogens. *Annu. Rev. Phytopathol.* 25:67-85.
- , AND R. J. COOK. 1974. Biological control of plant pathogens. American Phytopathological Society, St. Paul, MN.
- BRUCE, A., AND B. KING. 1983. Biological control of wood decay by *Lentinus lepideus* (Fr.) produced by *Scytalidium* and *Trichoderma* residues. *Mater. Org.* 18:171-181.
- , AND ———. 1986a. Biological control of decay in creosote treated distribution poles. I. Establishment of immunizing commensal fungi in poles. *Mater. Org.* 21:1-13.
- , AND ———. 1986b. Biological control of decay in creosote treated distribution poles. II. Control of decay in poles by immunizing commensal fungi. *Mater. Org.* 21:165-179.
- , W. J. AUSTIN, AND B. KING. 1984. Control of growth of *Lentinus lepideus* by volatiles from *Trichoderma*. *Trans. Br. Mycol. Soc.* 82:423-428.
- ESLYN, W. E. 1970. Utility pole decay. Part II: Basidiomycetes associated with decay in poles. *Wood Sci. Technol.* 4:97-103.
- GRAHAM, R. D. 1973. Preventing and stopping internal decay of Douglas-fir poles. *Holzforchung* 27:168-173.
- , AND M. E. CORDEN. 1980. Controlling biological deterioration of wood with volatile chemicals. Final Report, Project 212-1. Electric Power Research Institute, Palo Alto, CA.
- HULME, M. A., AND J. K. SHIELDS. 1972. Effect of primary fungal infection upon secondary colonization of birch bolts. *Mater. Org.* 7:177-188.
- KALLIO, T. 1971. Protection of spruce stumps against *Fomes annosus* (Fr.) Cooke by some wood-inhabiting fungi. *Acta For. Fenn.* 117. 20 pp.
- . 1972. The effect of *Gliocladium deliquescens* on the decaying capacity of some decay fungi. *Ann. Agric. Fenn.* 2:320-322.
- MORRELL, J. J., AND M. E. CORDEN. 1988. Evaluating potential decay control agents with a small block test. *Wood & Fiber Sci.* 20(4):479-486.
- MORRIS, P. I., D. J. DICKINSON, AND J. F. LEVY. 1984. The nature and control of decay in creosoted electricity poles. 1983 *Br. Wood Preserv. Assoc. Annu. Conv.*, pp. 42-53.
- NILSSON, T. 1973. Studies on wood degradation and cellulolytic activity of microfungi. *Stud. For. Suec. Nr.* 104.
- PAPAVIZAS, G. C., ED. 1981. Biological control in crop protection. Allanheld Osmun, London.
- . 1985. *Trichoderma* and *Gliocladium*: Biology, ecology, and potential for biocontrol. *Annu. Rev. Phytopathol.* 23:23-54.
- PRESTON, A. F., F. H. ERBISCH, K. R. KRAMM, AND A. E. LUND. 1982. Developments in the use of biological control for wood preservation. *Proc. Am. Wood Preserv. Assoc.* 78:53-61.

- RICARD, J. 1976. Biological control of decay in standing creosote treated poles. *J. Inst. Wood Sci.* 7(4):6-9.
- , AND W. B. BOLLEN. 1967. Inhibition of *Poria carbonica* by *Scytalidium* sp., an imperfect fungus isolated from Douglas-fir poles. *Can. J. Bot.* 46:643-647.
- ZABEL, R. A., F. F. LOMBARD, AND A. M. KENDERES. 1980. Fungi associated with decay in treated Douglas-fir transmission poles in the northeastern United States. *For. Prod. J.* 30(4):51-56.
- , C. J. K. WANG, AND F. C. TERRACINA. 1982. The fungal associates, detection, and fumigant control of decay in treated southern pine poles. Report EL 2768. Electric Power Research Institute, Palo Alto, CA.

#### WOOD SCIENCE AND TECHNOLOGY ACCREDITATION

SWST began accrediting schools for their Wood Science and Technology programs in 1984. The following is a list of schools accredited thus far.

<u>Program Reviewed</u>	<u>Year Accredited</u>
North Carolina State University, Raleigh	1984
University of Massachusetts, Amherst	1985
University of Washington, Seattle	1985
University of Minnesota, St. Paul	1985
Virginia Polytechnic Institute and State University, Blacksburg	1985
Mississippi State University, Mississippi State	1987
West Virginia University, Morgantown	1989