Auburn University Southern Forest Nursery Management Cooperative

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TREATMENT OF LONGLEAF PINE (*PINUS PALUSTRIS* MILL.) SEED WITH NINE DIFFERENT BIOLOGICAL AGENTS DOES NOT ENHANCE SEED GERMINATION

by
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INTRODUCTION

While the biological control of soil-borne pathogens has been studied for more than 70 years, the use of introduced microorganisms to control pathogens present in the soil rhizosphere has been a recent focus of biological control research in the forest industry. One reason for the increased use of biological agents in the forest industry is the phasing out of methyl bromide as a fumigant in 2001. Biological agents applied to seed prior to sowing in either bareroot or containerized nurseries could be used to not only control seedborne diseases caused by *Fusarium* but increase seedling biomass. Seed treatments could be used to control diseases caused by seedborne *Fusarium* sp. Currently, over-sowing longleaf pine seed in containerized nurseries is a common practice as up to four seeds may be placed in one container cell to compensate for unreliable germination of low-viability seed and the threat of damping-off. Since longleaf pine has unreliable seed crops, the seed is in particularly high demand. Thus, oversowing depletes the reserves of longleaf pine seed which are needed in years when poor seed crops occur. Thus, seed treatments could also be used to more efficiently use scarce sources of longleaf seed. This research was conducted to determine if nine biological agents, some previously used on agronomic crops, could increase germination of longleaf seed of two different viabilities.

METHODOLOGY

Greenhouse Experiments

Longleaf pine seeds were sown into 5 x 8, 40 cell containers filled with Pro-mix®, a peat-based soilless medium. Treatments consisted of 4 racks of 40 seed each for a total of 160 seed. Three rates, 1x, 2x, and 3x the label rate were applied to seed along with a non-treated control. The normal target crop, 1x rate, and reported benefits of the biological control agents used in these trials are presented in Table 1. Four biological agents, T22®, Deny®, Epic®, and Kodiak®, were applied in powdered form prior to sowing. The other five biologicals, B42, GB47, INR7, SE34, and SE49, were applied as a bacterial seed drench from working plates after sowing. For the seed drench treatments, working plates were flooded with 15 mL of sterile distilled water (SDW), bacteria scraped off using a sterile loop, and poured into 150 mL of SDW, 75 mL and 50 mL representing the 1x, 2x and 3x rates, respectively. Sown longleaf seed were drenched with 1 mL of each bacterial suspension. Controls consisted of 1 mL of SDW applied to each seed. Each experiment contained five replications of each treatment, and each experiment was repeated two times. The number of seed germinated were recorded every other day for 8 weeks. Germination for each bacteria and rate were analyzed using an ANOVA, and compared to non-treated controls to determine significance.

Table 1. Trade name, species, target crop, 1x rate and reported benefit of biological agent used to treat longleaf pine seed in 1998.

Strain	Species	Target Crop	1x Rate	Benefit
B42	*Bacillus* spp.	----------------	1.0 x 10^8 cfu/mL	-------------
Deny®	*Burkholderia cepacia*	Fruits, vegetables	0.045 g	Control damping-off
Epic®	*Bacillus subtilis* GB07	Wheat, barley	0.0225 g	Disease control
GB47	*Bacillus subtilis*	Cotton, snap bean	0.01 g	Soil-borne pathogens
INR7	*Bacillus pumilus*	Cucumber, tomato	1.0 x 10^8 cfu/mL	Induce disease resistance
Kodiak®	*Bacillus subtilis* GB03	Wheat, barley	0.01 g	Control damping-off
Se34	*Bacillus pumilus*	Cucumber, tomato	1.0 x 10^8 cfu/mL	Induce disease resistance
Se49	*Bacillus pumilus*	Cucumber	1.0 x 10^8 cfu/mL	Induce disease resistance
T22®	*Trichoderma harzianum*	Potato	1.0 x 10^8 cfu/mL	Damping-off prevention

Seed Sources

Six different seed sources representing three high-viability, and three low-viability seed sources were treated with each of the nine biological agents. Reported pre-trial percent germination ranged from 17-53% for low-viability seed sources and 92-95% for high-viability.

RESULTS

The effect of biological agents on longleaf pine seed germination can be grouped into two categories based on seed viability.

Biologicals and High-viability Seed Sources

The nine biological agents applied to seed sources with high-viability, B42, Deny®, T22®, Epic®, Kodiak®, GB47, INR7, SE34, and SE49 (Table 2), did not increase seed germination over pre-trial percentages, or over non-treated controls. Epic®, however, applied at the 3x rate significantly decreased the germination over non-treated controls, 51% versus 69%, respectively (Table 2).

Table 2. Percent germination of low-viability and high-viability seed lots 8 weeks post-treatment in greenhouse trials. Asterisk (*) indicates significant difference between treatment rate and control in a Dunnett's T-test (p=0.05).

	Low-viability				High-viability			
Biological/Species	0x	1x	2x	3x	0x	1x	2x	3 x
B42 (*Bacillus* sp.)	17	14	17	17	78	80	80	83
Deny® (*Burkholderia cepacia*)	18	19	21	13	73	64	74	62
T22® (*Trichoderma harzianum*)	19	27	14	18	85	77	75	77
Epic® (*Bacillus subtilis*)	47	49	44	36	69	59	61	51*
Bacillus subtilis	40	44	51	48	57	53	41	43
Kodiak® (*Bacillus subtilis*)	39	33	40	35	56	62	65	47
INR7 (*Bacillus pumilus*)	4	3	10	2	39	34	39	39
SE34 (*Bacillus pumilus*)	7	6	10	4	34	48	47	45
SE49 (*Bacillus pumilus*)	3	6	4	5	44	41	35	44

Biologicals and Low-viability Seed Sources

Treatment of low-viability longleaf pine seed with biological agents neither increased nor decreased the germination of longleaf pine seed. Longleaf pine germination for the individual treatments ranged from 3-51% for all three rates applied (Table 2).

MANAGEMENT IMPLICATIONS

The seed with low-viability was poor quality seed. Low germination percentages may have resulted because seed was empty or storage conditions have harmed seed. Treatment of the low-viability seed may not have increased the germination percentages. In the case of the high-viability seed the disease pressure may not have been high enough to produce results. Based on the experiments with the nine biologicals conducted in this research, the use of these on longleaf pine as a seed treatment to increase seed efficiency is not warranted.