

## ENHANCED SURVIVAL OF IMPATIENS IN A *RHIZOCTONIA SOLANI*-INFESTED MEDIUM BY BIOLOGICAL CONTROLS AND FUNGICIDES

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**Abstract.** A greenhouse experiment was conducted using 6-week-old plants of impatiens (*Impatiens wallerana* J. D. Hook) 'Dazzler Scarlet Plush' planted in styrofoam trays containing a peat-based potting medium. Biological controls [*Bacillus subtilis* (Ehrenberg) Cohn ( $1.2 \times 10^7$  cfu/ml), *Pseudomonas aureofasciens* Kluyver (63-28r,  $1.8 \times 10^7$  cfu/ml), or *Streptomyces violaceusniger* (Waksman and Curtis) Pridham et al. ( $5 \times 10^5$  cfu/ml)] were applied as soil drenches 14 days after planting (DAP). Fungicides [triflumizole (Terraguard 50 W, 0.3 g/l), PCNB (Terraclor 400, 0.47 ml/l), or fluazinam (500 F, 0.125 or 0.5 ml/l)] were applied as soil drenches at 20 DAP. Plants were inoculated by transplanting into a peat-based medium amended with *Rhizoctonia solani*-infested winter wheat seed in a ratio of 4:1 (w/w) and monitored for 1 month. Symptoms of *Rhizoctonia* crown rot were delayed and plant survival enhanced by *P. aureofasciens*, and *S. violaceusniger*, triflumizole, and low and high rates of fluazinam, but not by *B. subtilis* or PCNB.

Impatiens (*Impatiens wallerana*) has consistently been one of the most important bedding plants in the United States for the past 10 years (Niedbalski Cline et al., 1988). The fungus, *Rhizoctonia solani* Kuhn, affects a wide range of plant species and causes major losses in impatiens throughout the United States (Castillo and Peterson, 1990; Chase, 1991; Stephens et al., 1982; Niedbalski Cline et al., 1988) and in Australia (Harris et al., 1994b). Management of *R. solani* during bedding plant production currently involves the integration of fungicide applications and the use of pathogen-free propagative material, media, and containers (Stephens et al., 1983; Stephens and Stebbins, 1985; Harris et al., 1994b).

Interest in using bacterial and fungal biological control agents to reduce plant disease has increased due to increased regulatory constraints and heightened concern about nontarget environmental effects of pesticides (Campbell, 1989; McGovern, 1993). A number of bacteria have shown promise in reducing diseases incited by *R. solani* including *Bacillus subtilis* (Broadbent et al., 1971; Tschen et al., 1989), *Pseudomonas* spp. (Cartwright and Benson, 1995; Hagedorn et al., 1993), and *Streptomyces* spp. (Hwang et al., 1994; Rothrock and Gottlieb, 1985). Potential fungal biocontrols for *R. solani* include *Gliocladium*, *Trichoderma*, *Verticillium*, and binucleate *Rhizoctonia* spp. (Lumsden and Locke, 1989; Beagle-Ristaino and Papavizas, 1985; Van Den Boogert and Jager, 1984; Harris et al., 1994a). Although a number of these biocontrols have recently been commercialized or are under development, data are lacking concerning their efficacy on many crops including bedding plants.

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In addition to affecting impatiens production, *Rhizoctonia* crown and root rot is also a major limiting factor in the use of this popular plant in Florida landscapes. However, effective integrated management strategies have not been developed for *R. solani* in this setting. Recent reclassification of metam sodium as a restricted use pesticide has worsened this situation by leaving noncertified pesticide applicators, which includes the majority of homeowners, without suitable means for reducing inocula of the fungus. Some landscapers currently resort to the costly practice of soil removal and replacement at sites with high populations of fungus.

The purpose of this research was to evaluate the potential of biocontrols and fungicides at various stages of commercial development to delay symptoms and enhance the survival of impatiens in a medium infested with *R. solani*.

### Materials and Methods

A greenhouse experiment was conducted using eight impatiens 'Dazzler Scarlet Plush' plants per treatment or untreated control. Six-week-old impatiens seedlings were planted in styrofoam transplant trays comprised of 32-cm<sup>3</sup> cells, containing a peat-based medium (Metro Mix 220, Scotts) on 10 Mar. Ten ml of standardized suspensions of biocontrol agents *Bacillus subtilis* [UCC-0001, Uniroyal, Inc., ( $1.2 \times 10^7$  cfu/ml)], *Pseudomonas aureofasciens* [strain 63-28-r, Agrum Inc., Ag Biologicals, Saskatoon, Saskatchewan, ( $1.8 \times 10^7$  cfu/ml)] or *Streptomyces violaceusniger* [strain YCED-9, Actinovate Plus, Natural Industries, Inc., Dallas, TX ( $5 \times 10^5$  cfu/ml)] were applied as 10-ml soil drenches to plants on 24 Mar. (The viability and propagule count of each biocontrol were previously tested by growth following serial dilution on nutrient agar.) Actinovate has a broad label for use by plant growers as a growth-promoting soil amendment. *B. subtilis* is not currently available for ornamental use, although commercial formulations for reduction of soil-borne diseases of cotton exist. The biocontrol potential of *P. aureofasciens* is currently being evaluated on a number of ornamental crops.

Ten ml of triflumizole (Terraguard 50 W, 0.3 g/l), PCNB (pentachloronitro-benzene, Terraclor 400, 0.47 ml/l), or fluazinam (500 F, 0.125 or 0.5 ml/l) were applied as soil drenches on 30 Mar. Combinations of *B. subtilis* with either triflumizole or PCNB were also evaluated. Control plants were left untreated. Terraguard and Triflumizole are currently available for use by ornamental growers in the United States including producers of impatiens. Fluazinam is a relatively new pyridinamine compound with broad spectrum fungicidal activity (*Alternaria*, *Botrytis*, *Phytophthora*, *Plasmopara*, *Sclerotinia*, *Venturia*, etc.), and is labeled in the United States for use in on a variety of vegetable and fruit crops. This fungicide also demonstrated *in vitro* activity against *R. solani* and *Sclerotium rolfsii* Sacc. (Smith et al., 1992).

An isolate of *R. solani* which caused crown and root rot of New Guinea impatiens was grown in sterilized hard, red winter wheat seed and deionized water (1:1.5 w/v) for 1 month at 28C to produce inocula for the experiment. The infested wheat seed was then thoroughly incorporated into a peat-based potting medium (MetroMix 220, Scotts) using a 1:4 ratio (w/w) and placed in 15-cm (diameter) polystyrene pots.

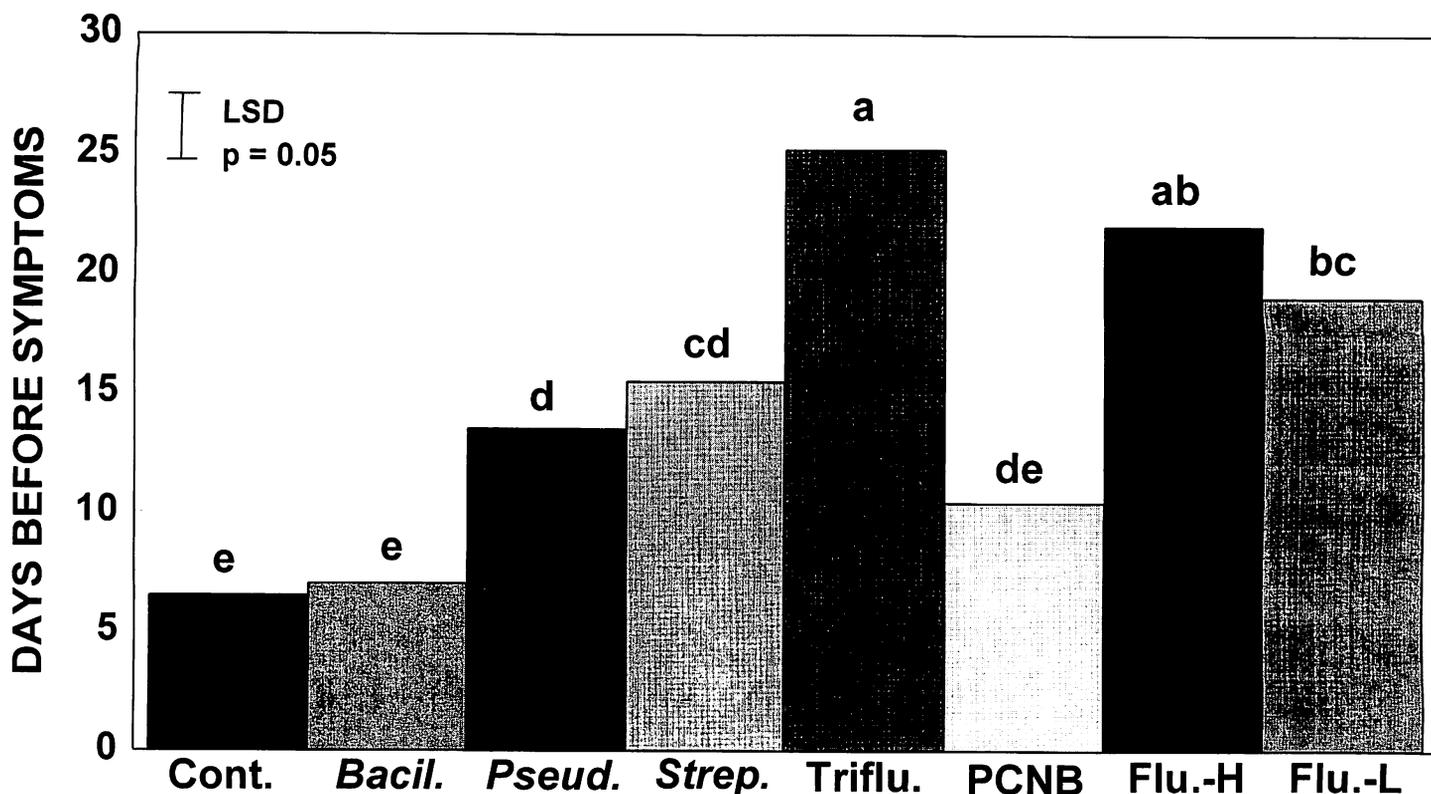


Figure 1. The effect of biocontrols and fungicides on the timing of symptoms of *Rhizoctonia* crown rot in impatiens 'Dazzler Scarlet Plush' planted in a peat-based medium infested by *R. solani*. Cont. = untreated control; Bacil. = *Bacillus subtilis*; Strep. = *Streptomyces violaceousniger*; Triflu. = triflumizole, PCNB = pentachloronitrobenzene; Flu.-H. and Flu.-L. = fluazinam high rate and low rate, respectively. Bars with different letters are significantly different at  $p = 0.05$  by LSD following ANOVA.

Impatiens were transplanted into the infested medium on 31 Mar. in an attempt to simulate the situation faced by landscapers at infested sites. Plants were arranged randomly on a bench and maintained in a greenhouse for 1 month at approximate maximum day/night temperatures of 38/28C, respectively, and fertilized daily with Nutrileaf 20N-8.8P-16.6K (20-20-20) water soluble fertilizer (Miller Chemical and Fertilizer Corp.) at a rate which delivered 50 ppm nitrogen. Plants were examined for one week to detect phytotoxicity, and every few days to determine the timing of disease appearance (crown rot, wilting) and length of survival. Treatment means were separated by the least significant difference test (LSD) following analysis of variance (ANOVA).

### Results and Discussion

No symptoms of phytotoxicity were detected as a result of any treatment. Rapid symptom expression and substantial mortality were observed in untreated 'Dazzler Scarlet Plush' (Fig. 1, 2). *Rhizoctonia* crown rot symptoms were significantly (LSD,  $p \leq 0.05$ ) delayed and plant survival enhanced by *P. aureofasciens*, *S. violaceousniger*, triflumizole and both rates of fluazinam; no plant death occurred where plants were treated with fluazinam at the highest rate. Symptoms of *R. solani* were not significantly delayed nor survival enhanced by either *B. subtilis* or PCNB, and neither synergistic nor additive effects were noted when *B. subtilis* was combined with triflumizole or PCNB (data not presented).

Stephens and Stebens (1985) found that PCNB was effective in controlling damping-off of impatiens by *R. solani*. Failure of the fungicide to suppress *R. solani* in the current

experiment may be due to the higher inoculum level used [multiple sources of inocula (wheat seed) dispersed through the planting medium of each plant vs. a single point source (agar disk) per row of seedlings], or the longer experimental duration (1 month vs. 1 week). Previous evaluations of *B. subtilis* as a biocontrol for *R. solani* have been variable; the bacterium was reported to control some isolates of the fungus (Broadbent et al., 1971; Tschern et al., 1989) while it failed to suppress others (Harris, et al., 1994b). Cartwright and Benson (1995) found that although *Rhizoctonia* stem rot of poinsettia was significantly reduced by *Burkholderia cepacia* (Palleroni and Holmes) Yabuchi et al. (synonym *Pseudomonas cepacia*), strains of other *Pseudomonas* spp. including *P. aureofasciens* achieved negligible reduction of the disease.

Additional research is necessary to confirm the results of the current study. Following such validation, the use *P. aureofasciens*, *S. violaceousniger*, and fluazinam may provide effective components of an integrated system for the management of *Rhizoctonia* crown rot of impatiens in both production and landscape sites.

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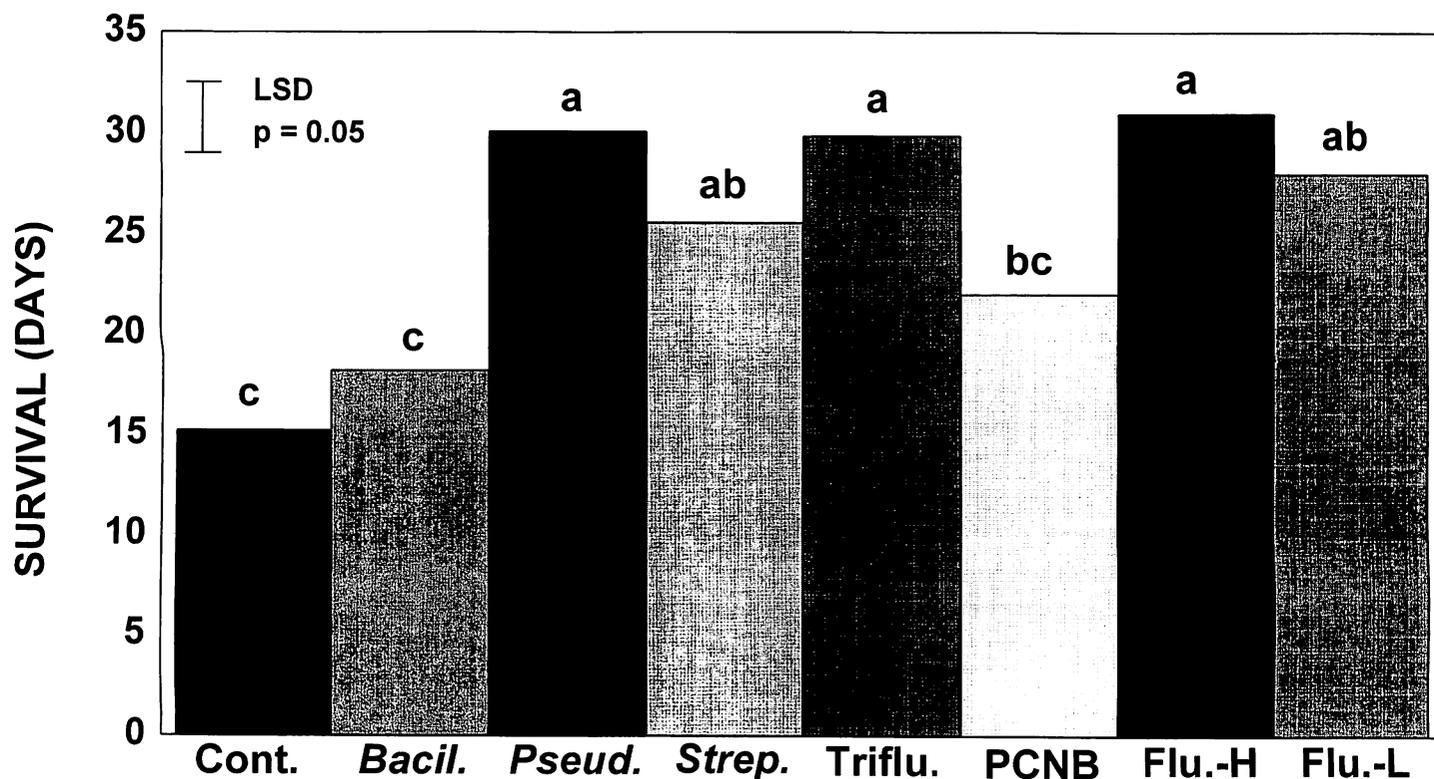


Figure 2. The effect of biocontrols and fungicides on the survival of impatiens 'Dazzler Scarlet Plush' planted in a peat-based medium infested by *R. solani*. Cont. = untreated control; Bacil. = *Bacillus subtilis*; Strep. = *Streptomyces violaceousniger*; Triflu. = triflumizole; PCNB = pentachloronitrobenzene; Flu.-H. and Flu.-L. = fluazinam high rate and low rate, respectively. Bars with different letters are significantly different at  $p = 0.05$  by LSD following ANOVA.

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