



Auburn University Southern Forest Nursery Management Cooperative

RESEARCH REPORT 12-06

CONTROL OF *RHIZOCTONIA* FOLIAR BLIGHT IN FOREST SEEDLING NURSERIES: A 3-YEAR STUDY.

by

Tom E. Starkey, Scott A. Enebak, Ken McQuage and Kevin Barfield

INTRODUCTION

The availability of fungicides to control specific forest seedling nursery diseases is either nonexistent, limited or faces possible loss of US label registration. Of the many insects and diseases that occur in forest-tree nurseries, three fungal pathogens stand out as problematic in southern US nurseries. These diseases include fusiform rust, pitch canker, and *Rhizoctonia* foliar blight. Nearly all nurseries in the southern US routinely apply fungicides to prevent fusiform rust on seedlings. *Rhizoctonia* foliar blight is the second most frequently occurring disease in this region.

Longleaf (*Pinus palustris*) and loblolly (*P. taeda*) pines are particularly susceptible to *Rhizoctonia* foliar blight (English and others 1986, Runion and Kelley 1993). The disease is caused by a species of *Rhizoctonia* spp. or binculeate forms of sexual states belonging to the genera *Thanatephorus* or *Ceratobasidium*. *Rhizoctonia* foliar blight can cause significant pine mortality in nursery beds and typically occurs in late July when the seedling canopy closes in (Carey and McQuage 2003). Symptoms of dead and dying needles and seedling mortality appear in patches within the bed where moisture and temperature favor infection. Often the disease is not observed until seedlings are top-clipped to maintain seedling shoot:root ratios and heights. Varying degrees of resistance among seedling families can be found, with US Gulf Coastal seedlots more susceptible than Piedmont sources, and the disease has not been observed on slash pine (*P. elliotti*) (McQuage, 2009 personal communication). *Rhizoctonia* foliar blight generally is not distributed uniformly throughout a nursery and is limited to foci within nursery beds. The disease is also more severe in second or third year crops post soil fumigation. While there are fungicides registered for *Rhizoctonia* foliar blight, they are not always efficacious (Carey and McQuage 2004) with azoxystrobin being the most commonly used fungicide (McQuage, 2009 personal communication). In most nurseries in the southern region, fungicide applications begin in mid-July when seedling canopy closure occurs.

In an attempt to find alternatives for the control nursery diseases, trials examining numerous fungicides by have been conducted by the Southern Forest Nursery Management Cooperative since 2004. In 2008, Proline[®] 480 SC (41% prothioconazole, Bayer CropScience), (Table 1), was examined as it had a broad spectrum systemic control of ascomycetes, basidiomycetes, and deuteromycetes on numerous field food crops. Prothioconazole belongs to the new chemical class of triazolinthiones (Mauler-Machnik and others 2002) and inhibits the demethylation process at position 14 of lanosterol or 24-methylene dihydrolanosterol, which are precursors of sterols in fungi. Prothioconazole efficiently stops many steps of the fungal infection chain like appressoria and haustoria formation, mycelial growth as well as spore formation. When these research studies were initiated, Proline[®] was only registered in the US for food crops including peanuts, barley, wheat, sugar beets, beans, soybeans and rapeseed. Beginning in 2008 data collected from the studies reported in this paper and other studies by the Southern Forest Nursery Management Cooperative were used to obtain a full-use label in December 2011 for disease control in forest-tree nurseries in the southern US.

METHODOLOGY

***Rhizoctonia* Foliar Blight Laboratory Trials**

Fungal growth studies were conducted in the laboratory to determine if *Rhizoctonia solani* was able to grow on agar media amended with Proline[®] at 1x, 0.25x and 0.0625x the label rate of 5 fl oz/ac. Potato Dextrose Agar (Difco[®] PDA) was amended with Proline[®] after autoclaving and just prior to pouring the plates. There were 20 PDA plates of each fungicide concentration and 20 non-amended PDA plates used as a control. A #4 cork-borer (8 mm) plug of *Rhizoctonia solani* taken from a 12-day old culture was placed at the center of each plate. The radial fungal growth was measured over a period of 7 days and recorded.

***Rhizoctonia* Foliar Blight Field Trials**

In 2008, Proline[®], at 5 fl oz/ac, and Abound[®] (23% azoxystrobin) at 24 fl oz/ac were tested at the Pearl River, MS nursery and was applied operationally for the control of *Rhizoctonia* foliar blight. A randomized block design with four replications was used in a nursery section growing its second seedling crop following soil fumigation. Each replication plot was 40 ft x 60 ft wide with a non-treated plot 20 ft x 60 ft left as the disease control (Figure 1). Seedling sample plots were assigned to bed-rows 2, 4, 6, and 8. Fungicides were applied on a two week interval beginning July 15, 2008 using a Hardee 1532 liter sprayer with a 9-bed spray boom; nozzles on 0.5 m centers. A total of 8 applications of both fungicides were made. Temperature and relative humidity were recorded just above the seed bed using a HOBO[®] data logger.

In early December 2008, seedling densities, disease incidence, severity and seedling loss were calculated in 2 subplots within each treatment plot. From each subplot, 30 seedlings were hand-lifted and, root collar diameter, height, dry weight and root morphology measured to determine seedling quality for each treatment.

In 2009, the identical study was established at the same nursery using the same experimental design and application methods. However, the fungicides were applied every 3 wk, to determine the minimal spraying time interval for disease control beginning mid-July.

In 2011, Proline[®] was tested at, 5 fl oz/ac and 3 fl oz/ac. Fungicide applications were made every 2 wk, beginning mid-July at canopy closure and applied operationally using the same experimental design described above. The purpose of this study was to determine if reduced rates of Proline[®] effectively controlled *Rhizoctonia* foliar blight. Seedling counts within each treatment were made at germination, prior to canopy closure and at prior to lifting to determine when seedling losses occurred.

RESULTS AND DISCUSSION

***Rhizoctonia* Foliar Blight Laboratory Trials**

Agar media amended with Proline[®] resulted in 100% control of *Rhizoctonia solani* as fungal growth did not occur on any of the Proline[®]-amended PDA plates for any concentration used for the 7 day experiment (Fig. 2). This control of fungal growth with all concentrations of Proline[®] cannot be directly extrapolated to field use, but it does indicate that rates lower than recommended label rates may be effective.

***Rhizoctonia* Foliar Blight Field Trials**

In 2008, when Proline[®] and Abound[®] were sprayed at label rates at two week intervals, disease incidence, severity and number of seedling mortality in Proline[®] treated plots was significantly less than in the Abound[®] and non-treated control plots (Table 2). The potential monetary loss due to *Rhizoctonia* foliar blight was \$967, \$209 and <\$1, per acre for non-treated, azoxystrobin, and prothioconazole, respectively. The potential monetary loss in Table 2 reflects the seedling loss in the test plot, not the whole nursery as *Rhizoctonia* foliar blight tends to occur in isolated foci in susceptible seedlots. There were no significant differences in either seedling quality or root morphology between the two fungicides tested, although the control plots had numerically fewer seedlings.

Seedlings sprayed with Proline[®] exhibited a “greening-up” effect that remained until the final application of Proline[®]. This “greening-up” effect had never been observed with any other fungicides at this nursery previously. Figures 3, 4 and 5 represent the control, Abound[®] and Proline[®] sprayed plots, respectively. The greening up effect has been observed on other crops treated with Proline[®] and it is reported that triazole fungicides can act to delay senescence by the generation of H₂O₂ as an intermediate metabolite (Audenaert and others 2010).

When the interval between fungicide sprays was increased from 2 to 3 wk, Abound[®] had a disease incidence of 8.6% compared to <1% for the Proline[®] (Table 2). When comparing the two studies, the disease intensity doubled for the Abound[®] applications and the potential loss per acre increased over 300% when applied every 2 wk versus every 3 wk. The potential monetary loss was \$1,294, \$693, and <\$1 per acre for non-treated, azoxystrobin, and prothioconazole, respectively. When using a fungicide other than Proline[®], it may not be possible to increase the spray interval greater than what is specified on the label. This study suggests that the time interval between Proline[®] sprays using suggested label rates is not as critical as with Abound[®].

The goal of the disease management programs at this nursery within these susceptible seedlots would be to keep seedling mortality to less than 0.5% (McQuage, 2009 personal

communication). Proline[®] was effective in reducing seedling mortality due to *Rhizoctonia* sp. below the nursery disease threshold.

In 2011, the appearance of *Rhizoctonia* foliar blight in the nursery was limited to the control plots in our study and the field edges that received uneven fungicide applications due to stopping and starting of the sprayer. In addition, the appearance of *Rhizoctonia* foliar blight did not occur until early September which is 3-4 wk later than previous growing seasons. When comparing Proline[®] sprayed at 5 and 3 fl oz/ac, more seedlings were lost at the 3 fl oz/ac than 5 fl oz/ac following canopy closure (Figure 6). After canopy closure the temperature and moisture conditions, along with seedling-to-seedling contact facilitated the aerial spread of *Rhizoctonia* foliar blight.

The appearance of *Rhizoctonia* foliar blight within the field plots in 2008 and 2009 was limited to the unsprayed control plot followed by plots treated with Abound[®] (Figure 7). There was little movement from either the control or Abound[®] plots into the Proline[®] plots. In 2011, *Rhizoctonia* foliar blight was limited to the control plots followed by the Proline[®] plots sprayed at 3 fl oz/ac. In each year the study-plot location was in a different area of the nursery. There was no correlation of disease spread and the prevailing wind direction during any year indicating that the inoculum within the field was the primary source for plot to plot and row to row spread (Figure 7).

Rhizoctonia foliar blight has 2 phases of disease development (Yang and others 1990). The first phase occurs before canopy closure, when the inoculum spreads within the field to seedlings by rain splash and irrigation. The second phase occurs after canopy closure when aerial spread of the fungus is facilitated by seedling-to-seedling contact in the presence of free moisture. Favorable fungal growth is reported to occur between 75° and 86° F (Copes and Scherm 2010, Frisina and Benson 1987) when there are 6 to 8 hrs of relative humidity > 95% (Copes and Scherm 2010). There is minimal fungal growth when the temperatures exceed 90° F (Frisina and Benson 1987).

Figures 8-10 show the environmental conditions over the growing season within each trial and indicate (red star) the time of occurrence of *Rhizoctonia* in the nursery. The shaded boxes are the period when the temperature and free moisture fell within the bounds favorable for *Rhizoctonia* reported above. The earliest occurrence of *Rhizoctonia* occurred in 2008 and the latest was in 2011. In 2011, it is possible that the aerial spread on *Rhizoctonia* was inhibited due to the high temperatures that summer. The temperatures in 2011 were the 3rd hottest temperatures on record in 117 years. Temperatures exceeded 90° F 69% of the days from canopy closure to the first appearance of *Rhizoctonia* foliar blight.

In December 2011, Proline[®] was approved by EPA for use in forest-tree nurseries. Proline[®] can be used to control diseases on nursery seed and seedlings of shortleaf, loblolly, slash and longleaf pines in addition to other pines, conifers and hardwoods. Before using any new pesticide, it is always a safe practice to apply it to a test area before larger areas in the nursery. The Proline[®] label can be viewed at <http://www.cdms.net/LDat/ld89K019.pdf>.

MANAGEMENT IMPLICATIONS

Laboratory studies have shown Proline® is capable of controlling fungi *in vitro* at rates much lower than the label rate of 5 fl oz/ac. In field studies, Proline® effectively controlled the spread of *Rhizoctonia* using 5 fl oz/ac which is within the range of 2.5 – 5.7 fl oz/ac for registered crops. Proline® provided significantly better control than Abound® when applied at a spray interval of every two or three weeks. Increasing spray intervals with fungicides other than Proline® may not be feasible in nurseries prone to *Rhizoctonia* foliar blight

The current practice of initiating fungicide applications for the control of *Rhizoctonia* foliar blight at canopy closure needs to be changed. Nurseries should start applying 1-2 fungicide applications before canopy closure when it is easier to apply fungicides to the lower stem and needles of the seedling. This will help to reduce the initial amount of inoculum found within the nursery beds and decrease disease pressure later in the growing season. In soybeans 60% of the primary infection of *Rhizoctonia* foliar blight occurred before canopy closure (Yang and others 1990).

Mulching may also help prevent initial infection within a nursery bed. In dry beans, (*Phaseolus vulgaris* L.), mulching was highly effective for the control of *Rhizoctonia* web blight (Galindo and others 1983). The mulched layer acted as a physical barrier to prevent or reduce the splashing of the inoculum up onto the bean tissue.

The aerial spread of the fungus depends upon free moisture in the canopy. Therefore, in nurseries where *Rhizoctonia* is a problem, irrigating early in the day so as to allow the foliar to dry before nightfall will reduce the available number of hours of free moisture.

Rhizoctonia foliar blight overwinters on seedling debris in the field. Use caution when sowing susceptible families on 2nd or 3rd year land following fumigation.

REFERENCES

- Audenaert K, Callewaert E, Hofte M, De Saeger S, Haesaert G. 2010. Hydrogen peroxide induced by the fungicide prothioconazole triggers deoxynivalenol (DON) production by *Fusarium graminearum*. BMC Microbiology 10:112 14 p.
- Carey WA, McQuage K. 2003. Evaluating fungicides for control of *Rhizoctonia* foliage blight of loblolly. Research Report 03-04. Auburn University Southern Forest Nursery Management Cooperative. 6p.
- Carey WA, McQuage K. 2004. Control of *Rhizoctonia* Blight by fungicides and fumigation. Research Report 04-3. Auburn University Southern Forest Nursery Management Cooperative. 4p.
- Copes WA, Scherm H. 2010. *Rhizoctonia* web blight development on container-grown Azalea in relation to time and environmental factors. Plant Disease 94: 891-897.

- English JT, Ploetz RC, Barnard E.L. 1986. Seedling blight of longleaf pine caused by a binucleate *Rhizoctonia* solani-like fungus. Plant Disease 70: 148-150.
- Frisina TA, Benson DM. 1987. Characterization and pathogenicity of binucleate *Rhizoctonia* spp. from Azaleas and other woody ornamental plants with web blight. Plant Disease 71:977-981.
- Galindo JJ, Abawi GS, Thurston HD, Galvez G. 1983. Effect of mulching on web blight of beans in Costa Rica. Phytopathology 73: 610–615.
- Mauler-Machnik A, Rosslenbroich HJ, Dutzmann S, Applegate J, Jautelat M. 2002. JAU 6476 - a new dimension DMI fungicide. The BCPC Conference Pests and Diseases 389-394.
- Runion GB, Kelley WD. 1993. Characterization of a binucleate *Rhizoctonia* species causing foliar blight of loblolly pine. Plant Disease 77: 754-755.
- Yang XB, Snow JP, Berggren GT. 1990. Analysis of epidemics of *Rhizoctonia* aerial blight of soybean in Louisiana. Phytopathology 80:386-392.

Table 1. Fungicide rates, active ingredient and manufacturer.

Fungicide	Active Ingredient	Manufacturer
Proline [®] 480 SC	prothioconazole 41%	Bayer CropScience
Abound [®]	azoxystrobin 23%	Syngenta Crop Protection

Table 2. Seedling density and disease loss as measured by incidence, severity and seedling loss per ft² and potential loss per acre caused by *Rhizoctonia* foliar blight in 2008 and 2009.

2008	Seedlings per square foot	Disease Incidence¹	Disease Severity²	Seedling loss per square foot	Potential Loss per Acre
TRT					
Control ³	22.9 (0.81)	0.089 (0.03)	0.3637 (0.14)	0.74	\$967
Abound [®]	23.6	0.041	0.167	0.16	\$209
Proline [®]	23.7	0.0008	0.003	0.00006	<\$1
<i>Prob > F</i>	0.7762	0.0004	0.0004	-	-
2009	Seedlings per square foot	Disease Incidence	Disease Severity	Seedling loss per square foot	Potential Loss per Acre
TRT					
Control	16.8 (0.92)	0.13 (0.03)	0.43 (0.11)	0.99	\$1,294
Abound [®]	20.5	0.086	0.30	0.53	\$693
Proline [®]	19.7	0.003	0.001	0.00006	<\$1
<i>Prob > F</i>	0.32	0.0044	0.007	-	-

¹ Incidence = proportion of RFB infected seedling per square foot of bed.

² Severity = proportion of tissue infected with RFB per square foot of bed.

³ Controls were not included in the statistical analysis due to lack of replication among blocks. Number in parenthesis is standard error of the mean.

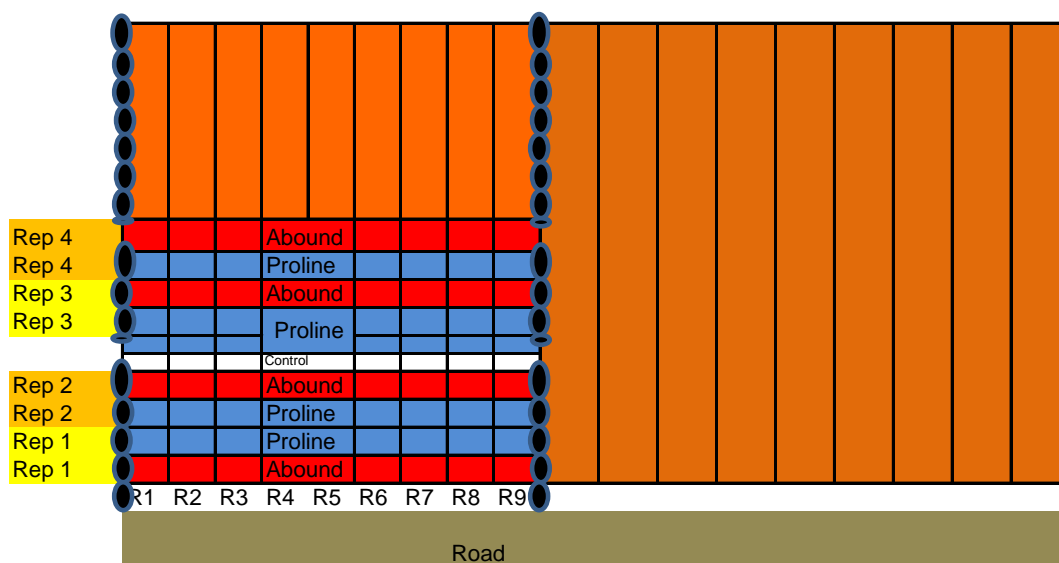


Figure 1. Experimental plot design used in each study 2008, 2009 and 2011.

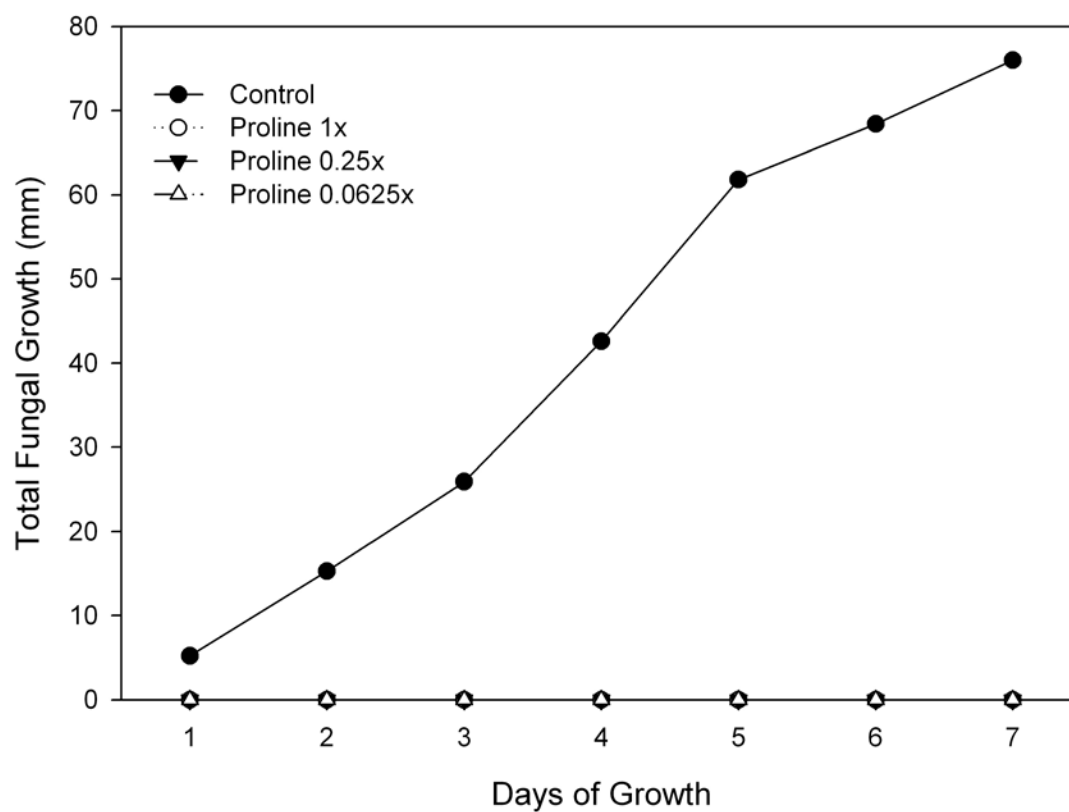


Figure 2. Radial growth of *Rhizoctonia solani* on fungicide-amended and non-amended media.



Figure 3. Control plot photo taken late September showing *Rhizoctonia* foliar blight.



Figure 4. Abound[®] plot photo taken late September showing *Rhizoctonia* foliar blight.



Figure 5. Proline[®] plot photo taken late September showing inhibited senescence and greening effect.

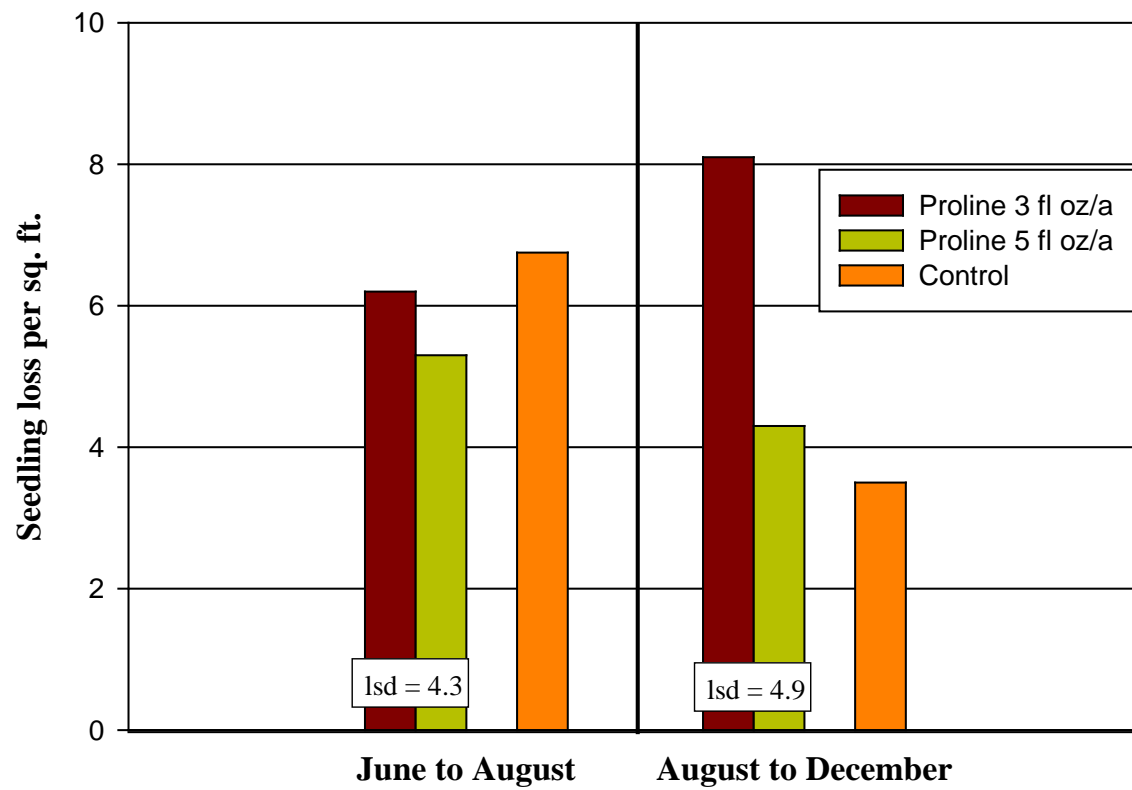


Figure 6. Loss of seedlings before canopy closure (June to August) and after canopy closure (August to December).

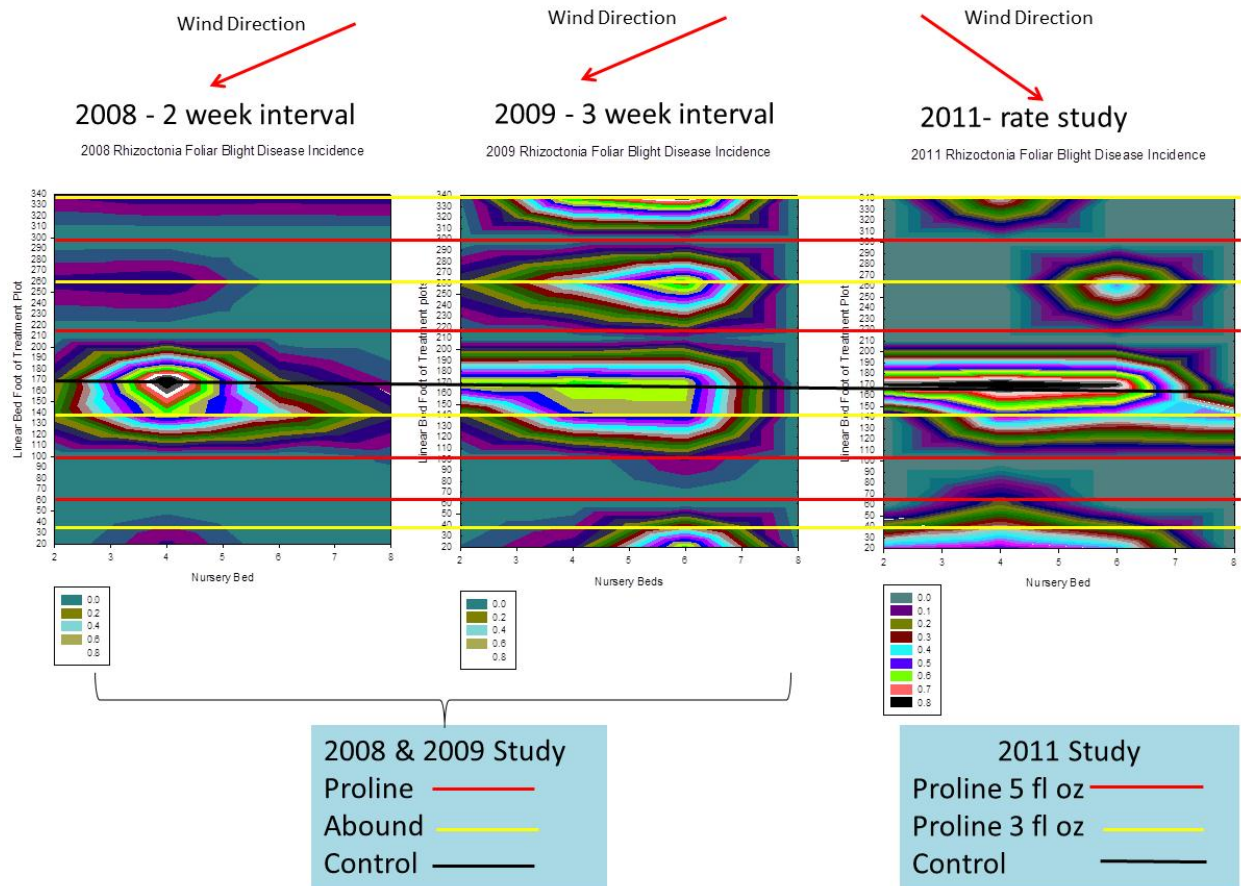


Figure 7. Disease incidence within nursery fields by year.

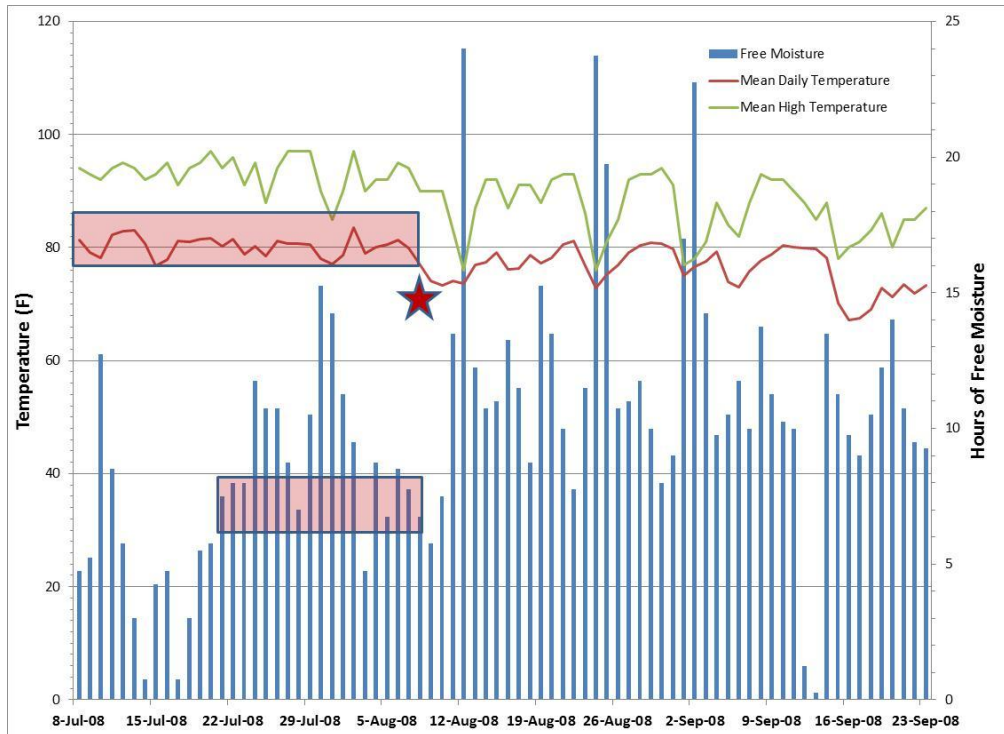


Figure 8. 2008 environmental data showing temperature, free moisture, date first *Rhizoctonia* foliar blight observed (star) and period in which environmental parameters favored fungal growth (shaded boxes).

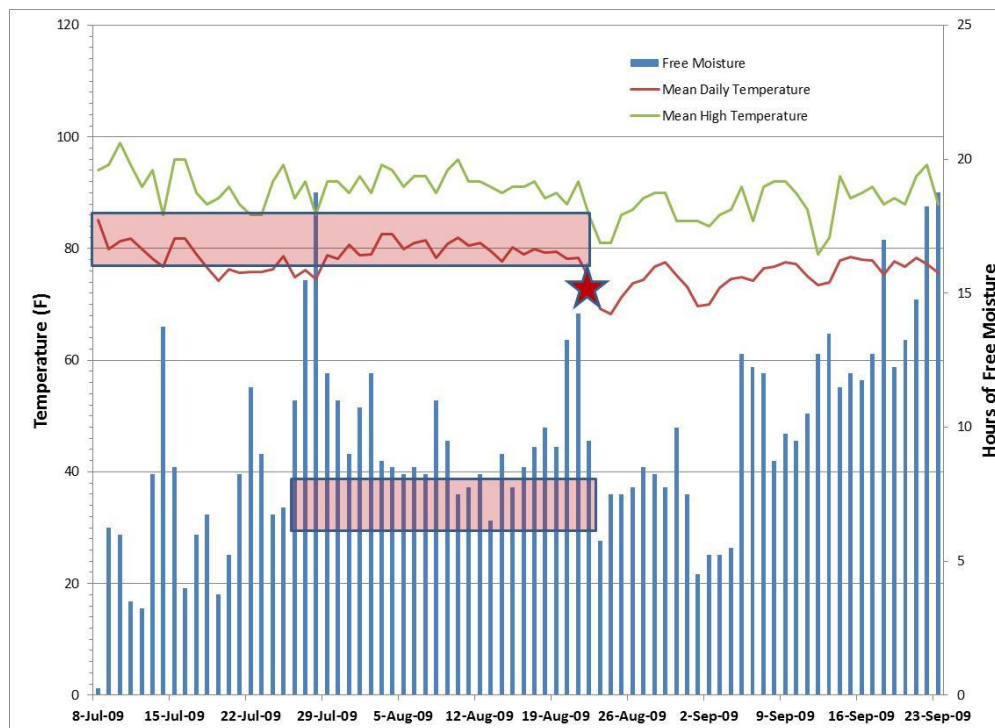


Figure 9. 2009 environmental data showing temperature, free moisture, date first *Rhizoctonia* foliar blight observed (star) and period in which environmental parameters favored fungal growth (shaded boxes).

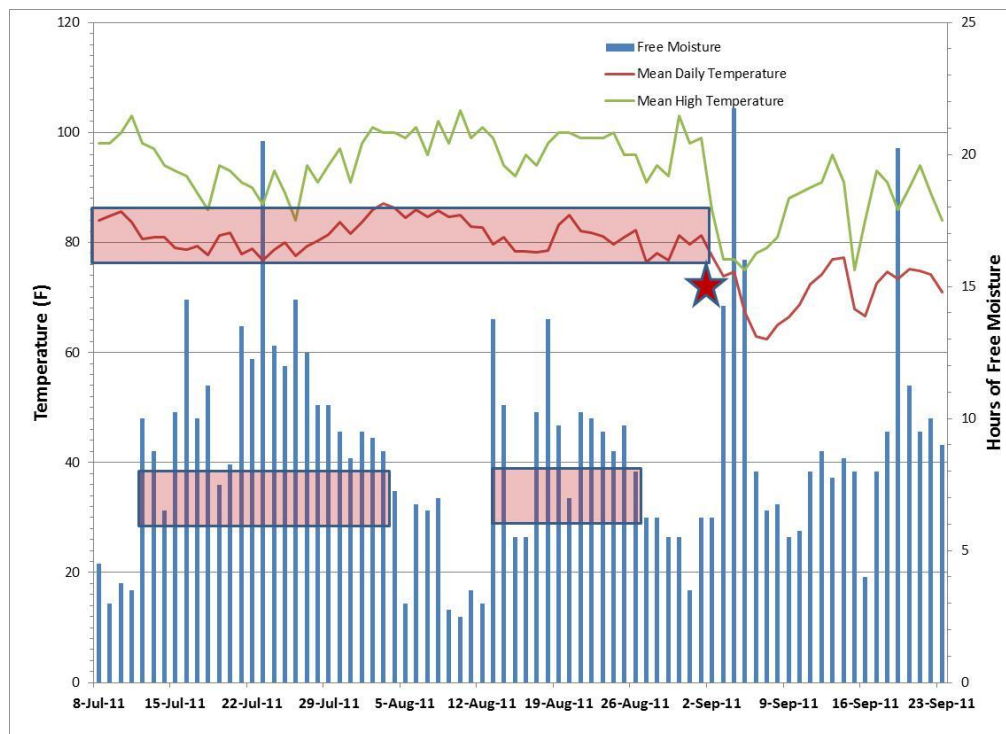


Figure 10. 2011 environmental data showing temperature, free moisture, date first *Rhizoctonia* foliar blight observed (star) and period in which environmental parameters favored fungal growth (shaded boxes).