

Soil Moisture and Fusarium Root Rot of White Pine Seedlings¹

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Abstract - Field and controlled environment studies were conducted to determine the influence of soil moisture on the development and severity of Fusarium root rot of white pine seedlings grown in sandy loam soil. During the 2+0 year in a Wisconsin nursery field, disease levels were significantly higher in two outer bed-rows closest to the irrigation line than beds closer to the center of the field. Soil moisture levels were also significantly higher in the outer bed-rows. Higher disease levels were positively correlated with wetter soils, and were observed in saturated and dry soil treatments compared to field capacity treatments in white pine seedlings grown in soil artificially infested with *Fusarium oxysporum*, untreated field soil, and pasteurized soil in leach tubes in a growth room study. Results suggest that prolonged exposure of white pine to saturated or dry soil conditions promotes Fusarium root rot. Options for managing soil moisture in forest nurseries are discussed.

INTRODUCTION

Soil moisture plays a major role in incidence, development and control of forest tree nursery disease and insect problems. According to Sutherland and Anderson (1980) excess soil moisture favors damping-off and root rot, particularly those caused by water molds (*Pythium* and *Phytophthora*), and is a common problem in bareroot nurseries with irrigation systems. Other types of damping-off and root pathogens such as *Rhizoctonia* are favored by moisture deficient soils.

Soil texture (size of soil particles) greatly influences the moisture holding capacity of soil. The ideal nursery soil is a sandy

loam that contains 15 - 20% silt and clay particles and 75 - 80% sand (Wilde, 1958). Ideally, nursery soils should be well drained.

In general, Fusarium diseases are more important in dry than wet soil conditions, although exceptions do exist (Cook and Papendick, 1972). Multiplication and survival of Fusaria are favored in sandy soils which are better drained, hold less water and dry faster than clays. Onset and severity of Fusarium diseases of winter wheat in the Pacific Northwest are favored by low plant water potentials (< -32 to -35 bars) (Cook, 1981). Susceptibility of sugar pine to Fusarium hypocotyl rot, caused by *Fusarium oxysporum*, was not affected, however, by plant water deficits of the magnitude recorded in a nursery field (Brownell and Schneider, 1985). Root necrosis of 2-year-old Douglas-fir seedlings due to *F. oxysporum* in Oregon occurred most commonly in wet areas of nursery beds (Hamm, et al., 1987). *Fusarium roseum* root rot of 1 and 2-year-old seedlings (white and loblolly pine, Douglas-fir, Norway spruce) was reportedly triggered by wet weather

and poor drainage in a Delaware Nursery (Schwalm, 1972).

Root rot has been observed in white pine seedlings and transplants in bareroot nurseries with sandy loam soils in the north central states (USA) and in southern Ontario (Canada) for many years. Several *Fusarium* species obtained from diseased white pine seedlings from an Ontario and a Wisconsin nursery have recently been demonstrated to cause root rot (O'camb and Juzwik, unpub.). *Fusarium* species are considered facultative parasites, i.e. they are able to grow saprophytically on non-living organic matter but are also able to cause disease in living plants. The most pronounced effects of water stress on disease development are evident with diseases caused by facultative parasites (Schoenweiss, 1978).

In 1989 we hypothesized that white pine root rot in northern nurseries was caused by *Fusarium*, and disease development was favored by soil moisture extremes. The objectives of our recent studies were to determine the influence of soil moisture and plant moisture stress on the development and severity of

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Fusarium root rot of white pine seedlings in nursery field and controlled environment conditions. Preliminary results of those investigations are presented here.

MATERIALS AND METHODS

Field Study

Study site

The field trial was conducted between 1989 and 1991 in one-half of a nursery section (165 m x 7.6 m) at Wilson Nursery, Boscobel, WI. Soils in this section were naturally infested with *Fusarium* spp. Mean soil-borne population of *Fusarium* spp. was 2206 colony forming units(cfu)/g dry soil during 1991. The soil in the section is a sandy loam with pH range of 5.4 - 6.0, maximum water holding capacity of 21%, an average bulk density of 1.21 g/cm³, and an organic content of 1 - 2 %. Irrigation water is applied through overhead, fixed pipes on permanent risers with oscillating nozzles.

Crop history

The section had a history of root rot in previous white pine crops. The section was fumigated with methyl bromide - chloropicrin (MC-33) in mid-August 1989, following incorporation of a sudan cover crop. Four beds were formed and white pine seed sown in early October 1989. Stand counts and seedling assessments were made through the 1+0 year prior to commencement of this study. Root disease was first apparent in the crop in September 1990.

Experimental design

The half section was divided into three blocks (55 m x 7.6 m). Two 1.1 m² permanent sampling plots were established in each of the four bed-rows in each block to give a randomized complete block design.

Monitoring and assessment

The total number of seedlings (living + dead) was determined for each plot in mid-May, late July and mid-October of the 2 + 0 year. Shoots of dead seedlings were cut at the ground line after counting. Ten living seedlings were randomly selected and removed from each plot on each sampling date for root assessment. The seedlings were stored in plastic bags at 4° C until processed. Daily mean soil matric potentials (smp) were determined at 10 cm depth in plot soils during the 2 + 0 growing season using electrical resistance (gypsum) blocks and electronic dataloggers (Campbell Scientific Inc., Logan, UT).

In the laboratory, seedling roots were carefully washed and the root system rated for evidence of root rot: 1= apparently healthy; 2 = over 50% length of one secondary root was necrotic; 3 = root necrosis in lower third of primary root or > 50% of two or more secondary roots necrotic; 4 = root necrosis in middle third of primary root; 5 = root necrosis in upper third of primary root or total root system dying or dead. Fungal isolations were also made from the edge of necrotic root tissue of affected seedlings. Excised tissue was placed on a *Fusarium* selective medium (Nash and Snyder, 1962), petri dishes

incubated for 14 - 21 days at 24 C, and then examined for *Fusarium* spp..

Data analyses

Disease data were summarized categorically by disease incidence and severity rating for each sampling date and cumulatively. Daily soil matric potential readings were classified in the following categories: wet = > -0.25 bars; upper field capacity = -0.25 to -0.40 bars; lower field capacity = -0.40 to -0.65 bars; and dry < -0.65 bars. The number of days that readings occurred in the different categories was tabulated by month, by half of growing season, and cumulatively. Disease and soil moisture data were analyzed using standard contingency table methods (Fienberg, 1980). Log linear modeling of treatment effects was used to further understand the contingency tables used.

Controlled Environment Study

Experimental design and soil treatments

A factorial design involving three soil moisture treatments and three soil fungal treatments was used for the controlled environment study. Sandy loam soil (pH = 5.4; maximum water-holding capacity = 23%; organic content of 1- 2%) was obtained from a white pine field at St. Williams Nursery, St. Williams, Ontario, Canada. Following mixing, three quarters of the soil was divided into 2 kg batches and autoclaved for 50 minutes at 1 kg/cm² (15 psi) and 121° C; the remaining quarter was not treated and served as the non-pasteurized field soil treatment.

Ground inoculum of a pathogenic isolate of *Fusarium oxysporum* f.sp. pini produced as an inoculum cake (Miles and Wilcoxson, 1984) was added to one-third of the pasteurized soil (1 g inoculum/kg soil) and mixed well. Another third of the pasteurized soil was designated as sterile control soil. Over 100 20-cm-long leach tubes (Stuewe and Sons Inc., Corvallis, OR) were filled to 13 cm height with the different soils. All tubes were then topped to 18 cm height with the remaining pasteurized soil.

Growing conditions and moisture treatments

Eastern white pine seeds were surface sterilized in 10% NaOCl for 25 minutes, rinsed in running water for 24 hours, and stratified for 60 days at 4°C. Seeds were sown in the pasteurized layers of all leach tubes, covered with pasteurized silica grit, and the soil watered to saturation. The trays with the tubes were moved to a walk-in growth room set for 20 C day/ 18 C night temperature, 18 hour photoperiod, and 60-70% relative humidity. Three weeks after sowing, the germinated seedlings were fertilized with a 11-41-8 fertilizer at 75 ppm nitrogen. Subsequently, seedlings were fertilized weekly at 100 ppm nitrogen using a 20-8-20 fertilizer. The seedlings were grown for 7 weeks (after sowing) before the following soil moisture treatments commenced: 1) saturated watered after 2-4 days when the soil moisture content (smc) reached 17% or smp < -0.1 bar; 2) field capacity - watered after 4-6 days when the smp was around 11% or smp between - 0.1 and -0.3

bars; and 3) dry – watered after 11-18 days when smp was around 5%. Watering regimes were based on mean tube weights that were determined to correlate well with gravimetric determinations of smp. The oven dry weight of the soil (105° C for 48 hours) was the reference weight used for calculating smp (Thompson and Troeh, 1973). The soil moisture treatments continued through two drying cycles of the dry treatment.

Assessment and sample processing

Soil and plants were removed from 30 leach tubes per treatment following the soil moisture treatment period. Soil moisture content was determined gravimetrically to verify moisture level achieved. Root systems of all seedlings were washed, exam-

ined, and rated for evidence of root rot per method described in field study section.

Data analyses

Root rot data were summarized categorically by disease incidence and severity rating. The data were analyzed using standard contingency table methods (Fienberg, 1980).

RESULTS

Field Studies

The total number of seedlings (living + dead) in each plot in May 1991 of the 2 + 0 year in the Wisconsin nursery field ranged from 158 to 471 per 1.1 m² plot with a mean of 321. Cumulative seedling mortality occurring within the plots between May and October 1991 averaged 7.5% (SE=1.4%). Seedling mortality

Table 1. Average number of days bed-row soils were determined to be in different soil moisture categories during 2+0 growing season in the Wisconsin nursery field

Bed-row no. ^a	Soil moisture category ^b			
	wet	upper field capacity	lower field capacity	dry
1	71	59	8	4
2	66	63	9	4
3	49	60	15	18
4	52	60	13	17

^a Six monitoring plots per bed row.

^b Wet = > -0.25 bars; upper field capacity = -0.25 to -0.40 bars; lower field capacity = -0.40 to -0.65 bars; and dry = < -0.65 bars

varied among the bed-rows ($P < 0.0001$), and was related to bed-row location. The highest mortality occurred in the two bed-rows nearest the irrigation line (10.4% for bed-rows 1 and 2, compared to 3.5% in 3 and 4; $P < 0.0001$).

By the end of the 2+0 year, average root rot incidence was 30.4% (SE=4.3%). *Fusarium* spp. were recovered from necrotic areas on primary and secondary roots of 72% of the seedlings visually assessed to have root rot. Significant effects of location on disease level appeared by July 1991, and became more pronounced by October 1991. In October, incidences were highest in the two bed-rows nearest the irrigation line (bed-row 1 = 50% and 2 = 33.3%) compared to the more distant bed-rows (18.3% and 20% in bed-rows 3 and 4, respectively) ($P = 0.0002$). Disease severity also appeared to be

highest in bed-rows 1 and 2 in the July and the October 1991 assessments.

Mean daily soil matric potentials (smp) were determined between 20 April and 8 October 1991 with electrical resistance blocks. No differences in monthly mean soil matric potentials among plots were apparent for April, May, September, and October. Apparent differences were, however, noted in mean monthly readings for June, July and August. Based on cumulative readings for the growing season, soils in bed-rows 1 and 2 were saturated (smp > -0.25 bars) for a significantly greater number of days than those in bed-rows 3 and 4 (Table 1) ($P < 0.001$). Conversely, soil in bed-rows 3 and 4 were dry (smp < -0.65 bars) for a significantly greater number of days than the other 2 bed-rows ($P < 0.001$)

We are conducting regression analyses using soil matric potentials and root disease levels to better define the relationship between the two variables. Preliminary results show that higher disease incidence is positively correlated with less negative soil matric potentials (wetter soil) and lower incidence with more negative soil matric potentials (drier soil) over the range of soil moisture conditions encountered in the field (Fig. 1).

Controlled Environment Study

Mean *Fusarium* spp. levels expressed as cfu/g dry soil in the container soils in the growth room experiment based on an assay of 15 tubes/soil treatment were: 44 for pasteurized control treatment; 201,591 for the pasteurized + *F. oxysporum* soil; and 2,146 for the non-pasteurized field soil

Table 2. Number of white pine seedlings that exhibited root rot at the end of controlled environment experiment on disease-soil moisture interaction

Soil treatment	Moisture treatment	D1 ^a	Severity rating ^b			
			1	2	3	>3
pasteurized	saturated	9	21	4	2	3
pasteurized	field capacity	5	25	2	2	1
pasteurized	dry	7	23	2	4	1
pasteurized + fungus ^c	saturated	15	15	5	10	0
pasteurized + fungus	field capacity	8	22	3	4	1
pasteurized + fungus	dry	13	17	5	7	1
non-pasteurized field	saturated	14	16	4	8	2
non-pasteurized field	field capacity	6	24	2	4	0
non-pasteurized field	dry	11	19	4	7	0

^a Number of affected seedlings per 30 examined

^b The higher the rating the more severe the root rot. See materials & methods section for description of ratings.

^c Pathogenic isolate of *Fusarium oxysporum* f.sp. *pini*

treatments. Minimum soil moisture content reached during the drying cycles for each moisture treatment over the 33 day treatment period were: 17% with 14 drying cycles for saturated; 11% with 7 drying cycles for field capacity; and < 5.5% with 2 drying cycles.

Elevated disease incidence and severity levels were observed for seedlings in saturated and dry soil treatments compared to field capacity in all soil treatments (Table 2). Preliminary analyses on disease incidence data revealed a highly significant effect of moisture treatment on seedling disease incidence ($P = 0.009$); soil treatment effect on disease incidence was not as pronounced ($P = 0.067$).

CONCLUSIONS

Preliminary results of the field study suggest that soils that are saturated over a large number of days during the growing season promote greater *Fusarium* root rot development in white pine seedlings than soils that apparently dry out reasonably quickly after irrigation or rainfall. Saturated soil conditions were observed in bed-rows with the highest disease levels for > 46% of the days monitored in the 2+0 year while dry soil conditions were more common (25% of monitored days) in bed-rows with lowest disease levels. Dry soil conditions experienced during the growing season did not appear to be severe or long enough in duration to increase root rot development. Ideally, nursery soils should be maintained at field capacity for optimal growth

and vigor. The cause of the prolonged saturated soil in two bed-rows of the field study is not known. Impeded drainage due to compacted soil layers is suspected. However, irrigation patterns and practices also need to be evaluated.

Preliminary results of controlled environment studies showed both saturated ($\geq 17\%$ swc) and dry (< 5.5%) soil conditions favored *Fusarium* root rot development compared to field capacity conditions (11 -17% swc) for a sandy loam soil.

Management Implications

Either excesses or deficits of soil water may cause significantly higher seedling root rot in lighter or heavier soils in forest nurseries, depending upon the pathogens involved. Considerable control over root disease may be achieved through appropriate soil management actions or practices without chemical use. Actions suggested by Cooley, et al. (1985) for managing *Phytophthora* root rot of conifers in the Pacific Northwest include improving drainage and tailoring irrigation practices to minimize disease. Drainage can be improved in fields with histories of root rot and excess soil moisture by installing a sub-surface drainage system, using ditches to remove surface water, sloping fields (through grading) toward roads or other non-cultivated areas, subsoiling to break up compacted soil layers, and elevating beds in the fields. Modification of irrigation systems or practices may also provide considerable control of root diseases in forest nurseries. For soil-borne diseases favored by

excess moisture, practices may be modified to avoid saturating soils continually or soils over extended periods of time, to avoid over-irrigation, and theoretically to avoid alternating drying and saturating of soil (i.e. try to maintain field capacity conditions as much as possible). For situations favored by soil moisture deficit, adjust timing, placement, and/or amount of water delivered to fields by the irrigation system to promote optimal seedling growth. In both excess and deficit soil moisture situations, seasonal monitoring and maintenance of irrigation equipment is required to ensure optimal irrigation patterns which deliver amounts of water appropriate for the crop growing stage and existing weather conditions.

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LITERATURE CITED

Brownell, K.H. and Schneider, R.W. 1985. Delimitation of lesions of *Fusarium* hypocotyl rot of pine by soil microsite environmental determinants. *Phytopathology* 75:58-60.

- Cook, R.J. 1981. Water relations in the biology of *Fusarium*. In *Fusarium: Diseases, biology and taxonomy*. Eds. Nelson, P.E., Tousson, T.A. and Cook, R.J., The Penn. State Univ. Press, University Park, PA. pp. 236-244.
- Cook, R.J. and Papendick, R.I. 1972. Influence of water potential of soils and plants on root disease. *Ann. Rev. Phytopath.* 10:349-374.
- Cooley, S.J., Hamm, P.B. and Hansen, E.M. 1985. Management guide of Phytophthora root rot in bareroot conifer nurseries of the Pacific Northwest. USDA For. Ser., PNW. 12p.
- Fienberg, S. 1980. The analysis of cross-classified categorical data. Mass. Inst. Tech. Press, Cambridge, MA. 198p.
- Hamm, P.B., Kanaskie, A., Morgan, P. and Cooley, S.J. 1987. Root necrosis caused by *Fusarium oxysporum* on 2-year-old douglas fir seedling in Oregon. *Plant Dis.* 71:651.
- Miles, M.R. and Wilcoxson, R.D. 1984. Production of fungal inoculum using a substrate of perlite, cornmeal and potato dextrose agar. *Plant Dis.* 68:310.
- Nash, S.M. and Snyder, W.C. 1962. Quantitative estimations by plate counts of propagules of the bean root rot *Fusarium* in field soil. *Phytopathology* 52:567-572.
- Schoenweiss, D.F. 1978. Water stress as a predisposing factor in plant disease. In *Water Deficits and Plant Growth*. Ed. Kozlowski, T.T. Vol. V. Academic Press, New York. 323p.
- Schwalm, J.M. 1972. Root rot in seedlings triggered by wet weather and poor drainage. *Tree Planters' Notes* 24:18.
- Sutherland, J.R. and Anderson, R.L. 1980. Seedling disease and insect problems related to nursery soil conditions in North America. In *Proc. North American Forest Tree Nursery*. Eds. Abrahamson, L.P. and Bickelhaupt, D.H. pp.182-190.
- Thompson, L.M. and Troch, F.R. 1973. *Soils and Soil Fertility*. McGraw Hill Book Co., St. Louis, MO. 495p.
- Wilde, S.A. 1958. *Forest Soils: their properties and relation to silviculture*. The Ronald Press, New York. 537p.