

ENVIRONMENTAL FACTORS INFLUENCING AIRBORNE ERGOT ASCOSPORE CONCENTRATIONS IN PERENNIAL RYEGRASS SEED FIELDS IN THE COLUMBIA BASIN OF OREGON

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Introduction

Ergot is an important disease of perennial ryegrass and can be a persistent problem in seed production systems. The disease is caused by the fungal pathogen *Claviceps purpurea*, which has a very wide host range among grasses and grains in North America. The fungus infects flowers prior to fertilization and colonizes the ovaries, resulting in the production of sclerotia rather than viable seed. Sclerotia are the overwintering structures of the fungus and produce airborne ascospores that serve as primary inoculum the following growing season. Ergot infection and infestation cause economic losses at various stages of grass seed production, including direct yield loss due to the production of sclerotia instead of seed, costs associated with protective fungicide applications, seed loss during recleaning processes that are required to remove ergot sclerotia from infested seed lots, and rejection of certification.

Cool-season grasses are grown for seed in a wide range of climates in Oregon, including mild and moist conditions in the Willamette Valley, semi-arid high elevation deserts in central Oregon and the Columbia Basin, and high mountain valleys in northeastern Oregon. Although ergot is a continual problem for grass seed production in eastern Oregon, the incidence and severity of ergot epidemics in grass grown for seed can vary among and within growing regions and from year to year (Alderman, 1991; Alderman et al., 1996; Alderman et al., 1998). In some years, the timing of ascospore release by the fungus may not coincide with grass anthesis, which is the only period of host susceptibility (Walenta et al., 2010).

Previous studies have investigated the timing and aerobiology of ergot ascospore production in Oregon, especially in Kentucky bluegrass (Alderman, 1993; Walenta et al., 2010). In the Willamette Valley, ascospore release in the field was associated with rain events occurring 2 to 3 days prior and was not correlated with temperature or relative humidity (Alderman, 1993). In the semi-arid Columbia Basin of eastern Oregon, grass seed production fields are frequently irrigated, and air or soil temperatures may play more important roles in ascospore release than precipitation events. A further understanding of the environmental conditions that contribute to ergot

ascospore production in eastern Oregon grass seed production systems is needed and could provide information that can be used to predict ascospore release and improve the timing of fungicide applications.

The objectives of this study were to: (1) determine the seasonal timing and concentration of ergot ascospores in fields of perennial ryegrass grown for seed in the Columbia Basin of eastern Oregon; (2) identify environmental factors that contribute to ascospore production; and (3) develop a model that can be used to predict ascospore production events in perennial ryegrass fields located in the Columbia Basin.

Materials and Methods

Spore sampling

Two commercial perennial ryegrass seed fields in Umatilla County, OR were included in this study. Field #1 was planted with cultivar 'Pavilion', and field #2 was planted with cultivar 'Top Hat II'. Both fields were 125 acres in size and were planted in the fall of 2011. Field #1 was located at an elevation of approximately 890 feet elevation, and field #2 was located at 712 feet. The fields were subjected to similar cultural practices and irrigated on a regular basis during the growing season using center pivot irrigation.

Burkard 7-day recording volumetric spore samplers were used to collect airborne ascospores of *C. purpurea*. In 2012, a spore sampler was placed in field #1 from May 11 to July 8. In 2013, spore samplers were placed in field #1 from April 3 to June 19 and in field #2 from April 3 to June 15. Spore samplers were placed approximately 500 feet from the field border with the air intake orifice located just above the mature canopy height. Spore trap tapes were collected and replaced weekly and were processed and analyzed as described by Alderman (1993).

Environmental data collection

Weather data were compiled from the HRMO weather station in the AgriMet Northwest Cooperative Agricultural Weather Network located at the Hermiston Agricultural Research and Extension Center. Soil temperatures were recorded 2 inches below the soil surface. Daily and cumulative degree days were

calculated for both air and soil temperatures using a base temperature of 50°F and an upper threshold temperature of 77° F for degree day calculations. These values were based on previous incubation studies, which found that ergot sclerotia germination was inhibited by temperatures outside of this range (Mitchell and Cooke, 1968).

Statistical analyses

Data collected from both fields and seasons were combined for statistical analyses. Correlation analysis was performed to identify significant ($P < 0.05$) correlations between daily ascospore concentrations and environmental data collected from the HRMO weather station. Local regression, which does not assume a linear relationship, was used to identify trends in daily ascospore counts against environmental variables (Cleveland et al., 1988). Results from correlation and local regression analyses were used to visually identify upper and lower threshold values of environmental factors that were significantly associated with ascospore occurrence. An environmental favorability index (EFI) model that included maximum and minimum soil temperatures, daily soil degree days, and mean dew point was developed to predict spore occurrence. Point values were assigned based on the following environmental variable ranges: maximum daily soil temperature between 59 and 72°F = 1 point; minimum daily soil temperature between 57 and 70°F = 1 point; daily soil degree days between 11 and 20 = 1 point; and mean daily dew point between 37 and 50°F = 1 point. Zero points were accumulated for any variable with values outside the above ranges. Chi-square analysis was used to determine the predictive ability of each environmental variable and the EFI model.

Results and Discussion

Spore traps used to monitor daily ascospore concentrations captured 27 spores in field #1 on the first day of trapping in 2012 (May 11), so spore traps were placed in fields earlier (April 3) in the 2013 season. The first occurrence of spores in 2013 was on April 24 in field #1 and on April 30 in field #2, when accumulated air temperature degree days reached 198 and 255, respectively. Accumulated soil temperature degree days at first spore occurrence in 2013 were 68 and 100 for field #1 and field #2, respectively. The majority of spores were captured between May 1 and June 15, with fewer spores sporadically captured before and after this time period. In field #1, more than nine times as many spores were captured in 2013 (more than 56,000) than in 2012 (more than 6,000). More than 114,000 ergot ascospores were captured in field #2 during the 2013 season. Such high concentrations of spores represent

significant sources of primary inoculum that can be extremely difficult to control.

Significant correlations ($P < 0.05$) were observed between spore counts and the following environmental variables collected from the HRMO weather station: minimum ($r = 0.25$), maximum ($r = 0.26$), and mean ($r = 0.29$) air temperatures; minimum ($r = 0.45$), maximum ($r = 0.24$), and mean ($r = 0.33$) soil temperatures; daily air ($r = 0.24$) and soil ($r = 0.38$) degree days; mean dew point ($r = 0.30$); and evapotranspiration ($r = 0.18$). Precipitation was not a significant factor in this study ($P > 0.67$), likely due to the regular irrigation that is required to grow grass seed crops in the semi-arid Columbia Basin.

Local regression identified several trends that were used to identify upper and lower threshold values for environmental factors significantly correlated with spore production. Peak spore production was associated with minimum air temperatures between 41 and 54°F and with minimum, maximum, and mean soil temperatures between 57 and 72°. These results are consistent with previous studies concluding that the highest percentage of ergot sclerotia germination was observed at incubation temperatures between 50 and 77°F, with germination reduced at temperatures below 41°F and above 77°F (Mitchell and Cooke, 1968). Daily soil degree days between 11 and 20 and a mean dew point between 37 and 50°F were also associated with spore production. Overall, variables associated with soil temperature (minimum, maximum, mean, and daily degree days) appeared to be more associated with spore production than other variables.

The environmental factors used to predict ergot ascospore occurrence in this study included minimum daily air temperature between 41 and 54°F, maximum daily soil temperature between 59 and 72°F, minimum and mean daily soil temperatures between 57 and 70°F, daily soil degree days between 11 and 20, and mean daily dew point between 37 and 50°F. When used to predict the appearance of at least one spore, all environmental variables except minimum air temperature were significant predictors ($P < 0.05$), with variables ranging in accuracy from 60 to 83 (Table 1). Table 1 also shows that all environmental variables were significant when used to predict the occurrence of at least 10 spores per day (64 to 90% accuracy) or at least 100 spores per day (60 to 89% accuracy). Environmental variables based on soil temperatures provided the most accurate predictions of spore events. A cumulative EFI was developed that included maximum soil temperature, minimum soil temperature, daily soil degree days, and mean dew point thresholds.

A cumulative EFI value of 2 correctly predicted the occurrence of at least one spore with an accuracy of 82% and correctly predicted the occurrence of at least ten spores with an accuracy of 86% (Table 2). A cumulative EFI value of 3 predicted the occurrence of at least 100 spores with an accuracy of 91% (Table 2). These results suggest that predictive models can be a useful tool to predict ergot ascospore production in the Columbia Basin.

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Table 1. Predictive accuracy of environmental variables used to forecast the occurrence of at least 1, 10, or 100 *Claviceps purpurea* ascospores.

Environmental variable	Spores present/absent			≥10 spores present/absent			≥100 spores present/absent		
	Correct prediction	False positive	False negative	Correct prediction	False positive	False negative	Correct prediction	False positive	False negative
	------(%)-----								
Minimum air T = 41 to 54°F	60.3 ¹	16.9	22.8	64.0	21.2	14.8	60.3	29.1	10.6
Maximum soil T = 59 to 72°F	74.1	0.0	25.9	86.2	0.0	13.8	88.9	4.8	6.3
Minimum soil T = 57 to 70°F	82.5	3.7	13.8	84.1	9.0	6.9	81.5	16.4	2.1
Mean soil T = 57 to 70°F	77.8	0.5	21.7	90.0	0.5	9.5	88.4	7.4	4.2
Daily soil degree days = 11 to 20	74.6	0.0	25.4	86.8	0.0	13.2	89.4	4.8	5.8
Mean dew point = 37 to 50°F	67.7	15.4	16.9	69.3	20.6	10.1	66.7	28.0	5.3

¹Chi-square result was not significant at $P < 0.05$. All other chi-square values (not shown) were significant at $P < 0.05$.

Table 2. Predictive accuracy of an environmental favorability index (EFI) model used to forecast the occurrence of at least 1, 10, or 100 *Claviceps purpurea* ascospores.¹

Environmental Favorability Index (EFI)	Spores present/absent			≥10 spores present/absent			≥100 spores present/absent		
	Correct prediction	False positive	False negative	Correct prediction	False positive	False negative	Correct prediction	False positive	False negative
	----- (%) -----								
EFI ≥ 1	78.9	15.3	5.8	77.3	22.2	0.5	66.1	33.9	0.0
EFI ≥ 2	81.5	3.7	14.8	86.2	7.4	6.4	83.6	14.8	1.6
EFI ≥ 3	73.5	0.0	26.5	85.7	0.0	14.3	90.5	3.7	5.8
EFI ≥ 4	65.1	0.0	34.9	77.2	0.0	22.8	86.2	1.6	12.2

¹All chi-square values (not shown) were significant at $P < 0.0001$.