**TIMING OF OCCURRENCE OF ERGOT ASCOSPORES IN NORTHEASTERN OREGON**

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### Introduction

Ergot, caused by *Claviceps purpurea* (Fr.) Tul., is a persistent problem in Kentucky bluegrass in northeastern Oregon, with direct seed loss as great as 25%. Ascospores, produced during the spring about the time of flowering in grasses, infect the grass ovaries. Seed loss can occur directly from replacement of seed with sclerotia, or indirectly when the sugary, sticky “honeydew” stage, which precedes formation of sclerotia, clumps seeds and debris together and sticks to machinery during harvest. Additional seed loss occurs during seed cleaning when re-cleaning is required to remove ergot to meet certification standards.

Ergot severity can vary from year to year, but the cause of variation is not well understood. Understanding the nature of the variability is important in developing control strategies for ergot. The objectives of this study were to determine whether variation in the severity of ergot in Kentucky bluegrass among sites is related to the timing and/or number of airborne ascospores during flowering, when the unfertilized grass ovaries are susceptible to infection, and to evaluate the efficacy of timing fungicide sprays for ergot control.

### Methods

**Fungicide trials:** Fungicide trials were established in fields of Kentucky bluegrass cv. “Midnight II” known to be infested with ergot at Site 1 (2008) and Site 3 (2009 and 2010). A commercial package mix of azoxystrobin (0.62 lb a.i./g) + propiconazole (1.04 lbs a.i./g), formulated as Quilt® and registered for use on grasses grown for seed in Oregon, was applied at label rate (14 oz/a) and included 1% v/v stylet oil. Treatments in 2008 included: a single application at early heading; an application at early heading and early flowering; an application at early heading, early flowering, and post flowering; and untreated control. Plot size was set at 90 ft. x 1000 ft. to accommodate a commercial field applicator (with 90 ft. boom), swather, and combine. Sprayer volume was 16 GPA at early heading and 18 GPA at early and post flowering, at 30 psi. Plot size restricted treatments to 3 replications.

Plot design in 2009 and 2010 was changed to include a single application at early flowering and a single application 14 days later (post flowering) among the treatments, utilize small plot equipment and include 4 replications in a randomized complete block design. Each plot was 5 ft. x 30 ft. Fungicide treatments were applied with a hand-held CO2 sprayer with 5 ft. hand boom fitted with TeeFet TurboJet® 60-11003 nozzle tips, delivering a spray volume of 18 GPA at 30 psi. In all years, fertilization, weed/insect management, and irrigation were managed by the cooperating grower and followed common production practices for the area.

In 2008, 2009, and 2010, plots were swathed on July 12, July 15, and Jul 26, and harvested on August 1, July 31, and August 5, respectively. Samples collected during harvest were cleaned and seed yield and percent ergot contamination was determined by dividing weight of sclerotia by weight of cleaned seed + sclerotia and multiplying by 100. Grass seed yield and percentage ergot contamination were subject to ANOVA and means were separated by the LSD all pair-wise comparison test. Data were analyzed using Statistix 9 (www.statistix.com).

**Ascospore and pollen trapping:** In each year of the study, ascospores were monitored in two commercial fields of Kentucky bluegrass cv. “Midnight II” and “SR2100” (Site 2) using Burkard 7 day volumetric spore traps (Burkard, Rickmansworth, England). The traps were placed within fields in early May and removed before harvest. Spore tapes were prepared, processed, and examined as previously described. The number of *C. purpurea* ascospores and grass pollen were counted under a microscope at 100-400X and summed over 12:00 pm to 11:59 am each day to establish daily counts. Standard slides of *C. purpurea* ascospores and Kentucky bluegrass pollen were used during the examination process. Differentiation of pollen to grass species was not attempted.

### Results and Discussion

**Fungicide trial:** Ergot was not detected in the fungicide trial plots in 2008. A low level of ergot (<0.51%) occurred in 2009 and 2010, but there was no significant difference in the level of ergot among the treatments.

Periodicity of ascospore and pollen release: Most ascospores of *C. purpurea* were trapped between 1:00 and 8:00 a.m. with few to no spores trapped between noon and 5:00 p.m. The occurrence of ascospores during the late evening through early morning hours is consistent with previously published data. Fewest grass pollen were trapped at about 5:00-6:00 a.m., followed by small peak between about 8:00 and 11:00 a.m. In *Poa pratensis* cultivars where the flowers open at night or early morning there would be a period when there are open flowers with unfertilized ovaries at about the same time as highest airborne ascospores numbers, increasing the chance of *C. purpurea* infection of Kentucky bluegrass. The scarcity or lack of spores during the afternoon suggests that in the Grande Ronde Valley, grass species that flower in the afternoon would be at less risk than Kentucky bluegrass, which typically flowers at night or morning, depending on cultivar. Resistance to ergot infection follows host fertilization, and consequently, flowers that escape infection and are pollinated later in the morning would develop resistance to infection.

Timing of ascospore and pollen release: Ascospores were trapped from mid-May through mid- to late-June (Figure 1). Total ascospores trapped at Site 1 in 2008, 2009 and 2010 were 7599, 181 and 1137, respectively. At Site 2 in 2008,
2009 and 2010 total ascospores trapped were 1332, 106 and 734, respectively. The long period of spore release is consistent with previous ergot spore trapping studies. The long spore release period is likely a reflection of variation in the timing of germination of sclerotia, and ascospore production from germinated sclerotia, which can extend for weeks.

The pattern of ascospore release was similar and ascospore numbers among the three sites were more or less parallel. This suggests that environmental conditions were similar among sites. The appearance of ascospores was several weeks in advance of flowering in Kentucky bluegrass occurring as early as May 14 in 2009 and 2010. The early appearance of ascospores suggests that early flowering grasses, including early flowering weed grasses, could be at greater risk of infection. Grasses such as annual bluegrass (Poa annua L.), which can flower weeks earlier than Kentucky bluegrass, could provide a potential source of secondary inoculum. The risk of infection from early flowering grasses in wheat has been demonstrated. Weed grasses common in Kentucky bluegrass fields in the Grand Ronda Valley that could potentially contribute sclerotia to the field including: cheatgrass (Bromus tectorum L.) and witchgrass (Panicum capillare L.).

In 2009, grass pollen was trapped about a week earlier than in 2008 and 2010 (Figure 2). Similarly, observed flowering in Kentucky bluegrass was earlier in 2009 and corresponded to a faster accumulation of degree days (Figure 3) relative to 2008 and 2010. Plant development, including the beginning and duration of flowering in Kentucky bluegrass is presumed to be a function of temperature. Most of the ascospores were released before the start of flowering, resulting in the crop escaping infection. In 2009, a greater overlap between ascospores and pollen occurred, although few ascospores were present, and at numbers that were not likely sufficient to promote more than a low level of infection. Warm spring temperatures, which promote early flowering in Kentucky bluegrass could place the crop at greater risk for infection if there is greater overlap with ascospore occurrence. It may be possible to use a crop degree day model to predict the early flowering in Kentucky bluegrass.

It is not clear if the ascospores trapped at each site originated solely from within the fields included in the study, or from other fields or areas. The trapping of pollen in advance of flowering suggests immigration of pollen from surrounding fields. Ascospores are presumed to also be capable of immigrating into a field from sources outside the field. In an area such as the Grand Ronda Valley, where winds are common and wind direction variable, a single infested field could be a significant source of inoculum for surrounding fields. Additional studies would be needed to determine the relative contribution and significance of within field and outside field inoculum sources.

Susceptibility to ergot varies among cultivars, and ranges from highly susceptible to resistant. In a location such as the Grand Ronde Valley, in which many Kentucky bluegrass fields are within relatively close proximity, avoiding highly susceptible cultivars would be preferred. In highly susceptible cultivars, it is doubtful that fungicides alone would provide adequate control. Attempts to control ergot with fungicides have been met with mix results.

It is not clear to what extent secondary spread of ergot occurred in the plots as honeydew occurrence was not quantified. The potential for secondary spread depends on the period that seed heads emerge and the duration of flowering within seed heads. This can vary among cultivars. Cultivars in which most seed heads emerge about the same time and in which flowering duration is short will have the least risk for secondary spread, as there will be few newly opened and unfertilized flowers available for infection by the time that honeydew is produced. In cultivars with a longer flowering duration, secondary spread would depend on the number and movement of insects capable of transferring conidia from infected to uninfected, unfertilized flowers. It remains to be determined if irrigation washes honeydew off plants or contributes to secondary spread through rain splash.

**Implications for control:** From an ergot management perspective, cultivars highly susceptible to ergot should be avoided when possible, alternate hosts for C. purpurea should be strictly controlled both within and surrounding the field, and efficacious fungicides should be applied at the beginning of flowering. In addition, it may be possible to reduce the number of sclerotia in the field following harvest by post-harvest field burning. Currently, growers typically spray for powdery mildew in April to early May and for ergot early to mid June, depending on start of flowering, with a second application 14 days later. Study results suggest that monitoring the level of airborne ascospores prior to and during flowering in Kentucky bluegrass might provide a means to determine whether or not to apply up to two fungicide sprays for ergot control. On a broader level, a degree day model to predict early flowering in Kentucky bluegrass may have potential to predict years in which the crop may be at greater risk for ergot. Additional studies will be needed to establish the potential of these approaches as decision aides to eliminate one or both fungicide sprays for ergot control.

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Figure 1. Daily percentage of total ascospores trapped in Kentucky bluegrass cv. “Midnight II” (Site 1 and 3) and “SR2100” (Site 2) in 2008-2010.
Figure 2. Daily percentage of total pollen trapped at in KBG cv. “Midnight II” (Site 1 and 3) and “SR2100” (Site 2) in 2008-2010.

Figure 3. Cumulative degree days at Imbler, OR, 2008, 2009, and 2010, based on degree day data from Agrimet, Imbler, OR site.