Improving Our Understanding of Aphanomyces Root Rot of Alfalfa

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Abstract: Aphanomyces root rot, caused by the oomycete pathogen *Aphanomyces euteiches*, can seriously impact alfalfa establishment, and is an important disease of alfalfa in poorly-drained soils in Kentucky. Field research was established to determine the impact of fungicide seed treatments on plant population and final yields in a field with a history of poor drainage and root rots in 2020. One treatment, Rizolex + Apron XL, increased plant populations compared to the non-treated control and other seed treatments, but there was no treatment effect on yield. Foliar fungicide applications and cutting schedule were also tested to determine if foliar inputs could increase yield in fields with a history of poor drainage and disease. Our research indicates that foliar fungicide did not increase yield in treatments on a 30-day or 40 day cutting schedule. COVID-19 restrictions delayed our ability to conduct complete *A. euteiches* race surveys as planned, and the destruction of the University of Kentucky Research and Education Center by a tornado on December 10, 2021 made finishing this portion of the project impossible.

Introduction:

Aphanomyces root rot, caused by the oomycete pathogen *Aphanomyces euteiches*, can seriously impact alfalfa establishment, and is an important disease of alfalfa in poorly-drained soils in Kentucky. Aphanomyces root rot can kill or stunt alfalfa seedlings and disease-stressed plants are unable to out-compete weeds, leading to thin, weedy stands. *Aphanomyces euteiches* survives in soil for many years, even when alfalfa is not planted, meaning that once the disease is present, alfalfa will always need to be managed for Aphanomyces root rot. In addition to the longevity of the pathogen, *A. euteiches* can evolve and adapt to become more aggressive and damaging.

Fungicide seed treatments have been available for alfalfa for several years, but recently new products have been labeled, some of which are specifically designed to manage Aphanomyces root rot. New chemical seed treatment active ingredients such as pyraclostrobin

and tolclofos-methyl are now labeled for *A. eutieches*, however their activity against *A. eutieches* has not be tested in public trials in Kentucky. These treatments have been shown to have some efficacy in other states (Smith, et al. 2015), but climate and population differences may impact efficacy in Kentucky. Foliar fungicides can also be used to improve alfalfa yield, and are promoted for improving "plant health" even in the absence of foliar disease. Very few foliar fungicide experiments on alfalfa have been conducted in Kentucky, especially in fields with poor drainage and other stresses.

Currently, *A. euteiches* race 1 and race 2 are present in Kentucky. However, the distribution and frequency of these races is not well-known since the last comprehensive survey for *A. euteiches* was conducted from 1990-1992 (Vincelli et al., 1994). One *A. euteiches* sample from Lexington, KY was race-typed in 2001 and determined to be race 2 (Malvick and Grau, 2001), and it is assumed that race 2 is widely prevalent in Kentucky, although there is little research to confirm this.

The objectives of this research were to 1) Determine if new chemical active ingredients have efficacy against *A. eutieches* in field research trials in Kentucky; 2) Determine if foliar fungicides improve alfalfa yield in field research trials, and 3) Soil sample alfalfa fields in Kentucky and determine race structure of *A. eutieches* using a race bio-assay.

Materials and Methods:

Research plots were established at the University of Kentucky Research and Education Center (UKREC) in Caldwell County, KY in 2020. The experiment was a randomized complete block design with four replications. Plots were 6-ft wide and 20-ft long, and the center rows were used for evaluation. The previous crop was soybeans. The field was poorly drained and had a history of root rot. Field tillage, fertilizer, and weed control were managed according to standard practices. The alfalfa cultivar Algonquin was drilled on a 7-in. row spacing at a rate of 15 lb seed/A on 3 Apr using a modified plot-sized drill.

To determine the effect of fungicide seed treatment on Aphanomyces root rot, fungicide seed treatments (Table 1) were applied using a batch seed treater prior to planting. Alfalfa stand was measured in each plot on 4 Jun, at the second trifoliate growth stage, by arbitrarily selecting two 1 sq ft areas within each plot and counting the total number of alfalfa plants in each square. Counts for each plot were averaged before analysis. The center 5 ft of each plot was harvested with a small plot harvester on 18 Aug and wet yield was obtained. A single 0.5 lb subsample of each harvest plot was collected and weighed, oven dried for 3 days, and weighed again to determine dry matter yield. Neutral detergent fiber (NDF), and acid detergent fiber (ADF) were determined using near infrared reflectance spectroscopy. Data were subjected to a mixed model analysis of variance and means were separated based on least squares means test at the 0.05 significance level.

In 2021, foliar fungicides were applied to the trial to determine if foliar fungicide and cutting schedule impacted yield. After an initial cutting, the foliar fungicides Headline (6 fl oz/A) or Priaxor (4 fl oz) were applied to research plots when plants were approximately 6-8 inches tall. A non-treated control was included. Plots were cut at 30 or 40 days after fungicide application. The center 5 ft of each plot was harvested with a small plot harvester on 16 Aug and 25 Aug and wet yield was obtained. A single 0.5 lb subsample of each harvest plot was collected and weighed, oven dried for 3 days, and weighed again to determine dry matter yield. Neutral detergent fiber (NDF), and acid detergent fiber (ADF) will be determined using near infrared reflectance spectroscopy. Data will be analyzed and means separated based on least squares means test at the 0.05 significance level.

A survey to determine the current race structure of *A. euteiches* in Kentucky was impacted by COVID-19. Travel could not occur in 2020, and was limited in 2021. Because a formal survey could not be conducted, we requested samples from County Agents across the state. We received 4 samples to date, which were stored for testing. We also could not get specific greenhouse supplies to conduct assays in spring of 2021 due to supply chain issues. Our greenhouse has limited cooling capacity, so research cannot be conducted in summer months. Experiments were scheduled to resume in early 2022 using an established bio-assay. However, the destruction of the University of Kentucky Research and Education Center by a tornado on December 10, 2021 made finishing this portion of the project impossible. Both samples and facilities were lost.

Project Objectives and Corresponding Results:

Objective	**Results**
Determine if new chemical active ingredients have efficacy against *A. eutieches* in field research trials in Kentucky	Rizolex + Apron XL seed treatment increased plant populations over the non-treated check and all other seed treatments; however treatment did not affect yield, ADF, or NDF. Results published in APS Journal Plant Disease Management Reports: Report No. 15:ST001
Determine if foliar fungicides improve alfalfa yield in field research trials	Foliar fungicides did not increase dry matter yield over the non-treated check on a 30 day or 40 day cutting schedule.
Soil sample alfalfa fields in Kentucky and determine race structure of *A. eutieches* using a race bio-assay;	Unable to complete due to tornado damage to samples and facilities

Distribute management information on Aphanomyces root rot in diverse formats to Kentucky alfalfa farmers.	Field trial results published in PDMR. Extension publication "An Overview of Aphanomyces Root Rot" was published through the Crop Protection Network (multi-state Extension group): doi.org/10.31274/cpn-20220526-0

Results and Discussion:

The trial location experienced below average temperatures in April and May of 2020 which delayed emergence and impacted stand and plant growth. Alfalfa growth was slower than expected with only one harvest possible. Treatment significantly influenced stand. Rizolex + Apron XL resulted in a significantly greater plant population compared to other treatments and the non-treated control. Apron XL alone resulted in greater population than the non-treated control. Treatment did not significantly affect dry matter yield, acid detergent fiber or neutral detergent fiber.

Table 1. Impact of fungicide seed treatment on plant population, dry matter yield, acid detergent fiber (ADF) and neutral detergent fiber (NDF) of alfalfa in field research trials in Princeton, KY, 2020.

Treatment, formulation, and rate/cwt	Population (plants/A)[z]	Total dry matter yield (lb/A)	Acid detergent fiber (ADF)	Neutral detergent fiber (NDF)
Non-treated check	368,337 c	2408.8	37.0	45.8
Apron XL, 3.0 SL, 0.64 fl oz	580,692 b	2466.7	39.6	48.6
Stamina, 1.67 FS, 1.50 fl oz	477,237 bc	2397.3	36.4	44.9
Apron XL, 3.0 SL, 0.64 fl oz + Stamina, 1.67 FS, 1.50 fl oz	406,452 bc	2030.0	35.7	44.1
Rizolex,, 4.17 FS, 0.30 fl oz	542,577 bc	1993.2	35.0	43.6
Rizolex, 4.17 FS, 0.30 fl oz + Apron XL, 3.0 SL, 0.64 fl oz	803,937 a	2595.6	37.5	46.4
P=	0.0014	0.5638	0.0910	0.1163

[z]Column numbers followed by the same letter are not significantly different according to least squares means tests at the *P*=0.05 level.

Analysis of foliar fungicide trials indicate that on a 30 or 40 day cutting schedule, foliar fungicide applications did not increase dry matter yield. No noticeable foliar diseases were present in the trial, which may have impacted results.

Table 2. Impact of foliar fungicides on dry matter yield of alfalfa in a 30-day cutting schedule in field research trials in Princeton, KY, 2021.

Treatment, cutting interval, rate	Total dry matter yield (lb/A)
Non-treated check, 30 day	2552.5
Headline, 30-day, 6 fl oz/A	2582.9
Priaxor, 30-day, 4 fl oz/A	2256.7
Non-treated check, 40 day	2320.1
Headline, 40-day, 6 fl oz/A	2097.2
Priaxor, 40-day, 4 fl oz/A	2408.3
P=	0.6795

Although certain fungicide seed treatments improved populations over the non-treated check, ultimately there were no differences in yield. Foliar fungicides also had no impact on dry matter yield in 2021. Additional research may help determine if seed treatments are a good investment in fields with a history of Aphanomyces or other root rots, and if foliar fungicides provide benefits in the absence of disease. This information is of benefit to alfalfa producers however since it provides additional information on if and when fungicide inputs are needed. These experiments indicate that expensive fungicide inputs may not be needed and other management options should be explored.

Research Publication:

Wise, K.A., Anderson, N., Teutsch, C., and Raymond, C. 2021. Evaluation of fungicide seed treatments on alfalfa in western Kentucky, 2020. Plant Disease Management Reports. Report No. 15:ST001

Extension Publication:

Wise, K., Smith, D., Samac, D. 2022. An Overview of Aphanomyces Root Rot. Crop Protection Network Publication CPN-6001. doi.org/10.31274/cpn-20220526-0.

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