

A Technique for Diagnosing *Aphanomyces* Root Rot on Alfalfa Seedlings

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This assay, as well as procedures for a soil bioassay to detect root-infecting oomycetes in alfalfa, was described by Paul Vincelli, Brian Eshenaur, Paul Bachi and Bill Nesmith in the *Plant Diagnostics Quarterly* in the early 1990's and has come in handy several times this spring already. It is a variation on the "float incubation" technique used by many of us (NPDN Diagnostic Tip of the Month by G. Ruhl, March 2007; Brock, J.H. and G.H. Beard, A Simplified Technique for Recovering *Pythium* and *Phytophthora* from Infected Plant Tissue).

Aphanomyces euteiches is a soil-borne oomycete that attacks the tender feeder roots of alfalfa seedlings causing weakening and stunting of plants (see *Compendium of Alfalfa Diseases*, 2nd ed., APS Press, St. Paul, MN). Leaves may become chlorotic or slightly reddened, and often curl downward; however, infected plants do not tend to collapse



Figure 1: Stunted alfalfa seedlings with root necrosis.

as in damping-off. Established plants are usually not seriously damaged by this root rot pathogen, but seedlings can be severely stunted or killed (Figure 1). *Aphanomyces* root rot occurs under

saturated field conditions, often in low areas that remain wet. Root rots caused by other oomycetes (such as *Phytophthora megasperma* f. sp.

medicaginis) produce similar symptoms and occur under similar conditions.

It can be difficult to isolate *Aphanomyces* sp. from root tissue, but we have found that a float incubation technique using germinated alfalfa seedlings as baits works well as a rapid diagnostic technique for confirming its presence in symptomatic alfalfa seedling roots. To prepare the bait seedlings, germinate alfalfa seeds (choose any *Aphanomyces*-susceptible variety) in a lightly moistened paper towel rolled up in a plastic bag. Incubated at room temperature, bait seedlings will be ready to use in 2-4 days; if needed, baits can be held in the refrigerator for about a week in the paper towel.

Gently wash the roots of stunted alfalfa plants; surface disinfection with bleach is not necessary and may inhibit sporulation of *Aphanomyces*. Place discolored and rotting alfalfa feeder root pieces (and taproot sections, if symptomatic) into a petri plate along with several of the bait

Diagnostic Updates



Figure 2: Select necrotic roots and place in incubation plate.



Figure 3: Add bait seedlings and incubate in de-ionized water at room temperature.

seedlings and partially cover with de-ionized water (Figures 2 and 3). Incubate at room temperature. Examine plates directly under the compound microscope (instead of removing tissue and placing on a slide). I begin to look for the characteristic sporangia with encysted zoospores at the apex (Figure 4) after about 30 hours, but it may take several days of incubation. Check plates frequently as other organisms may begin to obscure the *Aphanomyces*. The same technique may be used to detect *Phytophthora* root rot in alfalfa seedlings, using *Phytophthora*-susceptible alfalfa baits.

Photos courtesy of Julie Beale and Sara Long, University of Kentucky Plant Disease Diagnostic Laboratory. 🌿



Figure 4: Sporangium and zoospores of *A. euteiches*.

Two Day
Workshop:

Tree Pests of the Great Plains

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Judy O'Mara, Kansas State University, Department of Plant Pathology

This summer the Great Plains Tree Pest Council (GPTPC) and the Great Plains Diagnostic Network (GPDN) will host a two day workshop on the key insects, mites, diseases and disorders that afflict trees and shrubs on the Great Plains. The "hands-on" sessions will cover the key diagnostic concerns of specialists; cankers and wood decays of trees, identification of needle diseases of conifers, and identification of insect borers and defoliators. The workshop will also feature presentations on emerald ash borer and thousand cankers disease.

Registration

Registration is \$30 and must be received by June 23, 2010. There are a limited number of seats so register early! The registration includes refreshments at breaks and the Tuesday evening dinner at Fort Robinson. Make checks payable to the "South Dakota State University" and send to:

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Questions?

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Hotel accommodations

The Best Western West Hills Inn will be holding a block of rooms until June 13, 2010. Please call (877) 432 - 3305. 🌿