STAGONOSPORA LEAF SPOT AND CROWN ROT RESISTANCE

Test accepted: March 1991 **Test updated:** June 2024
Pathogen: *Stagonospora meliloti* (Lasch) Petr. **Test authors:** Donald Erwin, David Gilchrist, Larry Teuber, and L. D. Satterlee

PLANT CULTURE

Greenhouse

Container..... Permanent sand benches for direct seeding, plastic pots, or Conetainers; method should permit deep vertical tap root growth to 25 to 35 cm
Media..... Fertilized sand or UC soil mixture (50% silt; 50% peat moss)
Temp/Light..... 21 to 27°C; 14 to 16 hour daylength
No. of Plants ..... 35 to 50 plants per replication
No. of Reps ..... 3 minimum
Culture ..... Control insect populations; supplemental fertilizer (0-10-10) added once a month with water applied from below the crown.
Other ..... Inoculate seed with *Sinorhizobium meliloti*; seed directly into sand bench or into Conetainer; place cones in the sand (permits water and nutrient control and avoids excess water at crown)

INOCULUM CULTURE

Source ..... Pure cultures are isolated from alfalfa stems or crowns onto acidified potato dextrose agar or dilute V8 juice agar (V8JA) amended with streptomycin (30 ppm); radial growth is slow (2 mm per day); incubate infected leaves or stems containing pycnidia in a moist petri dish for 48 hours and streak pycnidiospores onto plates
Storage ..... Pure cultures on V8JA slants at 4°C; transfer pycnidiospores (not mycelium) to maintain pycnidial type
Storage life..... 6 to 10 weeks; renew by pycnidiospore transfer to fresh medium (avoid mycelium)

INOCULATION PROCEDURE

Age of Plant ..... Earliest stage for reproducible infection using CA isolates is two-week regrowth on plants cut at first flowering (30 to 45 days)
Type of Inoc. ..... Pycnidiospore suspension from 10 to 14 day old V8JA plates that have been flooded with spores
Concentration.... 10^6 spores per mL +0.01% Tween 20 (wetting agent); blended water agar may increase adherence

METHODS

Foliar..... Spray foliage until just before runoff, allow foliage to dry prior to misting
Crown..... Inject spores (10^6 per mL) into upper tap root or pour inoculum on scalpel wounds to prevent escapes

INCUBATION

Location ..... For leaf spot inoculate in mist chamber or greenhouse with mist system for 48 to 72 hours at 20 to 22°C and 16 hour daylength
Spacing..... 2.5 to 5 cm apart
Length..... Rate foliar lesions at 14 to 21 days after inoculation; rate crown rot at 2 to 6 months

RATING

Foliar

1 Resistant..... Free of symptoms
2 Resistant..... Lesions on leaves and petioles but none on stems
3 Resistant..... Lesions on leaves and up to 25% of stem tissue
4 Susceptible..... Lesions on leaves and 25% to 60% of stem tissue
5 Susceptible..... Lesions on leaves and 60% to 100 % of stem tissue

Root and Crown

0 Resistant..... No necrotic tissue at inoculation site
1 Resistant..... Local superficial lesion
2 Resistant..... Necrotic tissue (reddish flecks) with 1 to 5 mm lesion
3 Susceptible..... Necrotic tissue 5 to 10 mm laterally and 10 to 20 mm down from inoculation site
4 Susceptible..... Necrotic tissue 10 to 20 mm laterally and 20 to 30 mm down from inoculation site
5 Susceptible..... Necrotic throughout, dead plant

CHECK CULTIVARS

	Approximate Expected Resistance (%)	Acceptable Range of Reaction (%)
Moderately Resistant		
UC 129*	25	20-30
Susceptible		
Moapa 69	5	0-10

*Seed of UC129 is available from USDA-ARS National Plant Germplasm System (NPGS) from the main seed storage facilities at Washington State University - Pullman. See this site for details: https://www.naaic.org/resource/checkseed.php.

DISTRIBUTION AND SEVERITY OF STAGONOSPORA CROWN ROT

▲ States in which the disease has been reported.

Stagonospora stem and crown rot, *Stagonospora meliloti* (Lasch) Petr.
(Click on the map above for a larger version.)

CORRELATION TO FIELD REACTION

Greenhouse selections made at UC Davis and evaluated at three field locations in CA ranked similarly under all conditions.

CULTURE OPTIONS AND RANGE OF CONDITIONS

Pure cultures are grown on V8 Juice Agar at 22 to 24°C for pycnidial formation, and at 10 to 16°C for mycelial growth. Streaking of pycnidial spores or pouring a spore suspension on V8 plates will produce a "lawn" of pycnidia in 10 to 14 days. Light enhances pycnidial production.

HELPFUL INFORMATION

Temperatures may vary 21 to 27°C but higher temperatures reduce severity. All seedling plants tested in CA have been highly resistant to infection prior to emergence of the first flower buds. Reliable rating of genotypes requires regrowth following the first flowering. Foliar and stem infection is highly correlated with crown and root infection.

ALTERNATIVE METHODS

Since foliar inoculation is easier, faster, reliable and closer to natural conditions it is the method of choice, but crown inoculation also can be done by uprooting young plants after cutting the foliage, rinsing the roots with water, soaking the roots and crowns in a spore suspension then replanting. The incubation period for this method is two to three months.

REFERENCES

1. Erwin, D.C., R.A. Khan, O.K. Ribeiro, and W.F. Lehman. 1987. Growth sporulation and pathogenicity of *Stagonospora meliloti* and selection for resistance to crown and leaf spot of alfalfa. Plant Dis. 71: 181 - 185.
2. Erwin, D.C., R.A. Khan, and W.F. Lehman. 1987. Registration of UC 129 alfalfa germplasm with moderate resistance to Stagonospora leaf spot and root rot. Crop Sci. 27:820.
3. Insert Compendium of Alfalfa Diseases and Pests, 3rd Edition. 2015. APS Press, St. Paul, MN. 138 pp.