

# LEPTO LEAF SPOT RESISTANCE

Test accepted: March 1991

Test updated: June 2024

Pathogen: *Leptosphaerulina briosiana* (Poll.) Graham and Luttrell

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## PLANT CULTURE

### Greenhouse

Container ..... Flats or tool carts, 31x62x7.5 cm

Medium ..... Not critical

Temp/Light ..... 18 to 24°C; daylength not critical light, intensity >1000 Mmol m<sup>2</sup>sec<sup>-1</sup>

No. of Plants ..... 20 to 25 per replication

No. of Reps ..... 4 minimum

Other ..... Inoculate with *Sinorhizobium meliloti* or fertilize as needed; insect control, none within 1 week of inoculation.

## INOCULUM CULTURE

Source ..... Infected leaves

Storage ..... 6 months; longer on silica gel

Temperature ..... 4°C

## INOCULATION PROCEDURE

Age of Plant ..... 6 to 8 weeks or 4 to 5 trifoliates

Type of Inoc. .... Sporulating V-8 juice agar plate cultures, 3 to 10 days old, 19 to 23°C, cw fluorescence >20µmol m<sup>2</sup>sec<sup>-1</sup>

Method ..... Cultures inverted 30 to 60 cm above plants, approximately one culture per 900 cm<sup>2</sup> of plant material, in place until 10 spores per cm<sup>2</sup> collected in trap slides; culture plates should be relocated every 15 to 30 min to ensure uniform coverage; plants are sprayed with water when plates are removed.

Length ..... 2 to 4 hours, variable

Conditions ..... 100% relative humidity

## INCUBATION

Duration ..... 48 hours at 100% relative humidity, 20°C

Location ..... Move from moist conditions to green house after leaves have dried slowly out of direct sunlight.

Measurement .... Type and size of leaf spot, usually 10 to 14 days after inoculation.

## RATING

1 Resistant ..... No spots

2 Resistant ..... Barely visible black pepper spots, 1 mm diameter or less

3 Susceptible ..... Spots >1 mm, with or without tan center, no halo

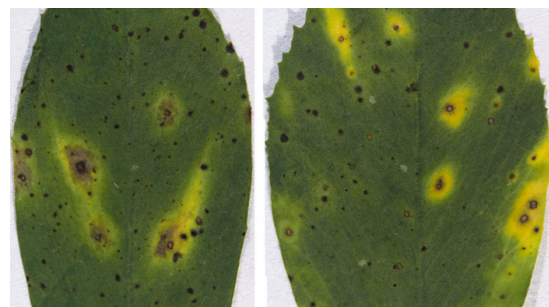
4 Susceptible ..... Spots with tan center, halo

5 Susceptible ..... Spots >3 mm, with tan center, halo, spots coalesced, leaf withered

## CHECK CULTIVARS

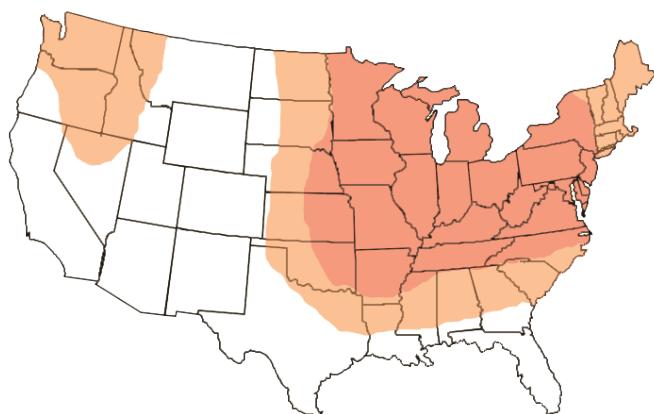
	Approximate Expected Resistance (%)	Acceptable Range of Reaction (%)
<b>Resistant</b>		
MSA-PL-L	25	
<b>Susceptible</b>		
Ranger	5	0-10
Moapa 69	5	0-10

Values for resistant standards are totals of 1's and 2's



(Click to see larger photo.)  
Symptoms of Lepto leaf spot.

## DISTRIBUTION AND SEVERITY OF LEPTO LEAF SPOT



- Not known to occur.
- Occurs but is not considered a problem.
- Occasionally causes significant losses on susceptible cultivars.
- Frequently causes significant losses on susceptible cultivars.

Lepto leaf spot, *Leptosphaerulina briosiana* (Poll.) Graham & Luttrell  
(Click map to the left for a larger version.)

## **CORRELATION TO FIELD REACTION**

Good correlations to field; no exceptions reported.

## **RACES**

No races are known.

## **CULTURE OPTIONS AND RANGE OF CONDITIONS**

Light is critical for spore production. Fluorescent or near UV is as effective as natural daylight. Some glass petri dishes do not pass sufficient light below 340 nm wavelength for good sporulation. Cultures can be started either by placing pieces of agar containing fungal hyphae onto agar surface or by spreading a spore suspension, (prepared by scraping surface of mature culture in water), over agar plate surface. The latter is the quicker method, but if plates contain bacterial contaminants they will not be usable. When cultures are ready to use, ascospores discharged onto petri dish lid are visible. These can serve as contaminant free source for subsequent cultures.

## **PLANT GROWTH OPTIONS AND RANGE OF CONDITIONS**

Vigorous plants are essential for expression of susceptible response. Light intensity after inoculation must exceed 1000 mole m<sup>-2</sup>sec<sup>-1</sup>. Supplemental light (metal halide or other) is necessary during winter at some locations to produce vigorous plants. Use of lightweight potting mix is best if plants are to be pulled during scoring.

## **INOCULATION CONDITIONS AND RANGE OF CONDITIONS**

Temperatures from 15 to 25°C are probably usable. Light during infection not required. Lower temperatures slow infection but seldom cause failure; too high temperatures or drying leaf surfaces will result in failure.

## **HELPFUL INFORMATION**

Plants may be cut back at scoring and regrowth used for different disease evaluation. Isolation of fungus is usually done by direct transfer of spores from sporulating pycnidia produced on leaf tissue. Infection does not kill stems or plants.

## **ALTERNATIVE METHODS**

Field evaluations may be possible but field infections are rarely of sufficient purity, uniformity, and severity to facilitate satisfactory selection. Inoculations have been made by spraying spores onto leaves. Cultures are scraped in water, comminuted, and filtered through cheesecloth to remove large particles. This method often results in a light inoculation.

## **REFERENCES**

1. Leath, K.T. 1971. Quality of light required for sporulation by *Leptosphaerulina*. *Phytopathology* 61:70-72.
2. Leath, K.T., and R.R. Hill, Jr. 1974. Large incubation chamber suited for use in selection for disease resistance. *Crop Sci.* 14:901-903.
3. Leath, K.T., and R.R. Hill, Jr. 1974. *Leptosphaerulina briosiana* on alfalfa: relation of lesion size to leaf age and light intensity. *Phytopathology* 64:243-245.