

# Root-lesion Nematode Resistance

*Pratylenchus penetrans* Cobb, Filipjev and Schur-Stekhoven  
D.K. Barnes, J.A. Thies, L.A. Wanschura and J.L. Townsend

## PLANT CULTURE

### Growth Chamber (Antibiosis) Test

Container ..... 3.8 cm x 19.0 polyethylene tubes (conetainers)  
Medium ..... Autoclaved 1:1 sand to soil mixture  
Temp/Light ..... 25 ° C; 16 hour daylength  
No. of Plants ..... 3 per tube (overplant and thin), 6+ replicates  
Other ..... Inoculate with *Rhizobium meliloti* Dang

### Field Screening Methods

Planting rate ..... 75 seeds per m of row or broadcast plots (1 m x 8 m plots) at approximately 55 viable seeds per 0.1 m<sup>2</sup>

## INOCULUM CULTURE AND PREPARATION

Source ..... Maintain *Pratylenchus penetrans* in monoxenic alfalfa callus (5) or in corn root explants at 25° C.  
Maintenance ..... Transfer alfalfa callus at 6 week intervals and corn root explant cultures at 3-4 month intervals.

## INOCULATION PROCEDURE

### Growth Chamber Test

Age of Plant ..... 12-14 days  
Inoc. Type ..... Suspension in tap water; extract nematodes from alfalfa callus or corn root tissue for 48 hours using shaker method (described in RATING section).  
Concentration ..... About 40 *P. penetrans* per mL  
Method ..... Inject 4 mL nematode suspension (150 nematodes per tube) into soil at 4 cm depth using a microliter pipette. Repeat inoculation 1 week later. Total = 300 nematodes/tube.  
Other ..... Include noninoculated control.

## INCUBATION

### Growth Chamber Test

Culture ..... Use insect free plants. Use of systemic insecticides is not advised; do not allow soil to become dry; avoid splashing of soil between tubes when watering. Clip plants 6 weeks after the second inoculation and evaluate after 4 additional weeks.  
Spacing ..... Place polyethylene tubes in alternate spaces in rack to allow for air circulation and ease of watering.

### Field Screening Method

Location ..... Field naturally infested with population densities of 3 or more nematodes per cm<sup>3</sup> soil.  
Culture ..... In severely infested soil, spray prepared seedbed with carbofuran (2,3-dihydro-2,2-dimethyl-7 benzofuranyl) methylcarbamate, flowable formulation) at 2.2 kg per ha a.i. before planting to allow plant establishment; based on Minnesota conditions, plots should be harvested 2X in the seeding year and 3X in the second year.  
Rating ..... In mid-September of the second year, plants are undercut and rated for root damage.

## RATING

### Growth Chamber Test

Shoot dry weight, fibrous root dry weight, tap root dry weight (not including crown), and numbers of nematodes within the roots are recorded. Nematode numbers within roots per tube are the most important data. Nematode numbers are obtained by cutting fibrous roots into 1 cm sections. A 1.5 g fresh weight sample is placed in a 10.0 cm x 2.5 cm petri dish containing 20 mL of distilled water, and placed on a horizontal shaker. After 7 days, water is decanted and the nematodes are counted using a stereomicroscope. Alternatives are pan extraction (7) and staining nematodes in the roots with acid fuchsin (2).

### Field Screening Methods

Resistant plants (scored 1 or 2) have a large amount of top growth, good crown development, and many fibrous roots with few lesions.

### Root Damage Score

1 Resistant ..... Normal, healthy root system with abundant fibrous roots  
2 Resistant ..... Small reduction in the amount of fibrous roots  
3 Susceptible ..... Moderate reduction in fibrous roots  
4 Susceptible ..... Total loss of fibrous root system as well as lesions on taproot  
5 Susceptible ..... Plant dead

## CHECK CULTIVARS

Resistant ..... MNGRN- 16 has an antibiosis level that supports about 60% fewer root lesion nematodes than Baker.  
Moderately Resistant ..... MNGRN-4 is tolerant and has a low level of antibiosis. Germplasm supports about 20% fewer root lesion nematodes per g fresh root weight than Baker (1).  
Susceptible ..... Baker

## DISTRIBUTION AND SEVERITY OF ROOT-LESION NEMATODE



Root-lesion nematode, *Pratylenchus penetrans* Cobb, Filipjev and Schur-Stekhoven (3,9).

Click on the map above for a larger version. See also the [KEY](#).

### SOURCE OF INOCULUM AND SEED

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### HELPFUL INFORMATION

Growth chamber and field studies (4,6) have both been used to characterize resistance. Because antibiosis and tolerance mechanisms are both important, comparing resistance among cultivars is difficult. Field procedures can be used effectively for selecting resistant germplasm but growth chamber tests are required for making claims about antibiosis resistance mechanisms. Nematode number data is inherently variable, thus requiring many replicates (6 to 15) to statistically separate entries. Seed of the resistant checks (MNGRN-4 and MNGRN- 16) can be obtained from T. A. Campbell.

### ALTERNATIVE METHODS TO COMPARE CLONES

#### (8)

Grow rooted clonal stem cuttings for 8 weeks in sterile soil. Transplant plants to 12.5 cm pots containing soil infested with approximately 1,000 *P. penetrans* per pot. Ten weeks later extract nematodes from each root system for 1 week in a mist chamber, and record as nematodes per g fresh root.

### REFERENCES

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