

TWO NEW TECHNIQUES FOR SCREENING PEAS FOR RESISTANCE AGAINST ASOCHYTA PISI

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For successful selection of peas resistant against Ascochyta pisi Lib. rapid and effective screening techniques are necessary. Two new techniques are described here. The seedling test is well suited for single plant selection, especially in breeding programs with semi-leafless plant types. The detached leaf test could be useful for simultaneous tests with different pathotypes as several leaves of the same plant can be used.

The tests were performed with 6 lines proposed as standard differentials by Darby, Lewis and Matthews (1): Dik Trom (JI 423), Rondo (JI 502), Wyola (JI 320), Frazer (JI 403), Pisum elatius (JI 198), Pisum jomardii (JI 250).

The following isolates were used for resistance tests:

British pathotypes 1-5 (1)

Dutch Race E

16 isolates collected at 8 different locations in Germany.

Cultures of the different isolates were grown on pea agar (extract of deep frozen peas 150 g/l, 5 g glucose/l and 12 g agar/l) at 20°C for 7 days. Inoculum was prepared by washing spores from cultures after 10 days of cultivation. Spore concentration was adjusted to 2.5×10^5 spores/ml. Tween 80 (0.05%) and milk powder (0.5%) were added in order to improve spore germination and to enhance viscosity of the suspension.

Seedling test

Pea seedlings in the two leaf stage were used for inoculation. About 0.1 ml of the spore suspension was applied to the apex with a large needle of a sterile syringe. Whereas inoculation with the spraying method (1) requires artificially raised high humidity, normal growing conditions are sufficient for the seedling test method. The humidity necessary for spore germination is available at the apex and maintained for some hours between the enclosing stipules. Escapes seem to be less frequent than with the spraying method.

The assessment of symptom severity can be made after two weeks. The classification is recorded on a scale proposed by Darby et al. (1):

- 0 Rare hypersensitive reactions
- 1 Hypersensitive reactions, slight wilt, no lesions or stem infection
- 2 Occasional small lesions, no stem lesions
- 3 General leaf and stipule infection, no stem lesions
- 4 General leaf and stipule infection, stem lesions
- 5 General leaf, stipule and stem infection, frequent death of seedlings

Frequently in lines of reaction class 1 development of primary branches could be observed without any lesions on the main stem. This seems to be due to a retarded growth at the apex and is related to the hypersensitive reaction observed in the detached leaf test.

Detached leaf test

Fully expanded leaflets or stipules of plants in the 4-6 leaf stage were harvested and the leaves placed in petri dishes containing tap water so that the lower end of the leaf reached into the water while most of the leaf floated on the surface on strips of Parafilm. Three to four small droplets (10 µl) of the spore suspension were placed on each leaf. The petri dishes were then incubated in a growth room at 20°C under an alternating cycle of 12 h light and 12 h darkness.

Disease symptoms were scored two weeks after inoculation. Three different types of reaction could be observed: a symptomless or sometimes hypersensitive reaction in the resistant lines, an intermediate reaction type with lesions of about 3-6 mm size with only a few pycnidia formed and a very susceptible reaction type with large lesions (> 10 mm) and a high level of pycnidium production.

For both tests a good congruity was obtained compared with the classification of Darby et al. (1).

Pathotype determination

In summer 1988, isolates were collected from different locations, mainly in North-Germany, to perform pathotype determination. Six of the 16 isolates tested could be identified as Race E, four each as British pathotypes 1 and 2 (corresponding to Dutch Race C), and one isolate each as British pathotypes 4 (Race D) and 5 (Race B), respectively. There was no isolate which did not correspond to one of the hitherto known races confirming the relatively stable pathotype situation in Ascochyta pisi as already discussed by Darby et al. (1).

J1-lines and isolates of the British pathotypes were kindly provided by Dr. P. Matthews, Norwich, and the isolate of race E by Dr. M. Gerlagh, Wageningen.

1. Darby, P., B.G. Lewis and P. Matthews. 1986. Plant Pathology 35: 214-223.
