

The testing of rape for clubroot
(*Plasmodiophora brassicae* L.)

KAREL BÍLEK

OSEVA, Breeding Station at Slapy, near Tábor town

Isolation, obtaining and preservation of pathogen

Inoculum of durable spores is obtained from galls of plants infected, and the obtainment just before plant disintegration is the best. Galls can be stored at the temperature -13°C , even in the course of several years without any loss of their viability. The viability is negatively influenced by thawing and then refreezing the material. Such an inoculum is also possible to obtain when sowing sensitive plants (Chinese cabbage cv. 'Granaat' is the most suitable) into the soil severely infested.

Inoculum preparation

An appropriate inoculum is prepared by homogenizing galls being fresh or frozen in distilled water. The homogenate obtained is filtered through a close plastic screen. Suspension is then adjusted with haemocytometer to the concentration required (to 10^6 spores/ml the most often). The suspension prepared in this way can be used even during two months provided that it is maintained at the temperature up to $+5^{\circ}\text{C}$.

Preparation of material

Thirty plants at least from every genotype are necessary to be evaluated. Only uniform seed being of good germinating capacity and untreated must be used for testing.

Inoculation

Growing substrate is prepared by mixing soil, peat and expanded perlite to be in ratio 2:4:1. Afterwards the substrate is sterilized with superheated steam for 30 min. Growing substrate obtained is filled into multipots the surface of which is smoothed. Then the substrate in individual pots is pressed in

order that five germinative seeds can be sown. By means of a doser 0,5 ml of inoculum is added to each of seeds. After inoculation seeds are covered up with perlite.

Incubation

Gall development is dependent on satisfactory moisture content in growing substrate as well as on its temperature (optimum temperature is 20 - 25°C). In winter period there is a need of supplementary illumination. When emerged the number of plantlets per pot is reduced to 3. After 10 days the temperature is possible to decrease to 18°C.

Evaluation

Evaluation is carried out within 6-7 weeks after inoculation. Individual plantlets are carefully pulled out, the substrate is removed from them, and then are rinsed in water so that galls arisen can be seen well.

Evaluation is made using the scale ranging from 0 to 3

- 0 - without infestation
- 1 - small galls on secondary roots
- 2 - medium-sized galls
- 3 - all or majority of roots modified into galls

Using values obtained the percentage of infestation is calculated, and even disease index (D. I.) is possible to determine. D. I. in sensitive genotypes is usually 80 - 100, and that in highly resistant genotypes is less than 20. Distinctness and/or agreement of disease severity between individual genotypes is expressed on the basis of statistical evaluation.