

A RAPID TEST FOR EVALUATING RESISTANCE TO SUNFLOWER STEM CANKER
CAUSED BY *DIAPORTHE/PHOMOPSIS HELIANTHI*

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SUMMARY

A rapid test has been developed for the assessment of resistance to *Diaporthe/Phomopsis* stem canker of sunflower under both controlled and semi-field conditions. Sunflower seedlings were produced either in a growth room or in field plots under plastic tunnel, wound-inoculated by the fungus at 6-leaf-stage, and incubated in moist conditions. Evaluation was made based on a disease rating 2 weeks from inoculation and a degree of resistance was calculated. The growth room test, although more severe, allowed us to differentiate better between susceptible /e.g. NSH-26, NSH-68/ and tolerant /NSH-43/ sunflowers.

INTRODUCTION

Stem canker caused by *Diaporthe helianthi* Munt.-Cvet. et al. /anamorph: *Phomopsis helianthi* Munt.-Cvet. et al./ is a disease of major importance in sunflower production, particularly in Hungary /Vörös et al. 1983/ and some other European countries /Acimovic and Straser 1981; Iliescu et al. 1983; Lamarque and Perny 1985/. It may be a limiting factor of this crop in other continents as well /Herr et al. 1983/.

Although chemical treatments are promising /Iliescu et al. 1983/, the production of sunflower with genetically controlled tolerance or resistance seems to be more reasonable. Thus, Vranceanu et al. /1983/ reported about a selecting work carried out under field conditions, and showed a great variability existing in sunflower inbred lines for their reaction to *D. helianthi* attack. Similarly, breeders at Novi Sad detected some lines carrying genes for resistance to stem canker, and have already released hybrids with such genes /Skoric 1985/. They also used a field selection and natural infection by the fungus. In a recent report, Bertrand and Tourvieille /1987/ compared different methods of inoculation and they found petiole-test as the most reliable for determining the level of resistance.

In order to get as rapid information as possible about the reaction of new sunflower cultivars to *Diaporthe* infection, a testing method has been devised for being less dependent upon seasonal variation. By using a standard inoculation technique and controlled environment, a series of NSH-hybrids were tested and the results compared to our field experience.

MATERIALS AND METHODS

Sunflower seedlings of the cultivars tested were grown either in pots on a growth room bench at $22 \pm 2^{\circ}\text{C}$, illuminated for 15 h per day /approx. 12 000 lux/, or they were seeded directly into field plots protected by a plastic tunnel. At 6-leaf-stage /approx. 5 weeks from seeding/ the seedlings were wound-inoculated by making a tangential cutting with a scalpel of each stem at the first internode, putting a mycelium disc /8 mm ϕ / from a 4-week-old

culture of the fungus on potato dextrose agar onto the wound, and dressing it with wetted cotton pad. Finally, the inoculated stem part was covered with aluminum foil to maintain a high level of moisture during incubation. At least 15 plants of each cultivar were inoculated and wounded, and non-inoculated wounded plants served as control. All the experiments were repeated twice. Disease incidence and severity were recorded by counting the number of seedlings with stem lesions and by determining the extent of both external and internal tissue necrosis 14 days from inoculation. Each plant was rated on a scale from 0 to 4 as follows: 0, nil; 1, lesion restricted to inoculation site and outer cortical tissues, stem hard; 2, stem slightly staved at inoculation site, lesion half around, localized tissue necrosis inside the stem; 3, lesion around the stem with definite staving, partial necrosis of pith; 4, thin and often broken stem with extended necrosis in and on the stem.

RESULTS

In a preliminary experiment, various disease parameters, as lesion size and colour, stem diameter and length, the number of healthy leaves, as well as the proportion of stem tissues showing necrosis in cross section were compared. As a result, it was concluded that internal tissue necrosis was the most suitable symptom for the evaluation, whereas all other parameters including lesion size gave inconsistent result. Consequently, our disease rating was primarily based on assessing the intensity of transversal stem necrosis further. To estimate the value of this testing method, a series of NSH-hybrids has been compared under both growth room and semi-field conditions. Unfortunately, the tests conducted so far under plastic tunnel did not provide with consistent result which is partly due to extreme weather during the season. The results of the growth room experiments are shown in Table 1. Although the infection rates were relatively high and significant differences could hardly be obtained between the hybrids tested, NSH-43 showed fairly good tolerance to *Diaporthe* infection, and NSH-68 was found to be as susceptible as the control hybrid.

Table 1 Reaction of NSH-hybrids to *Diaporthe* infection using wound inoculation in a growth room 5 weeks from seeding

Sunflower hybrid	Average disease scale /max. 4/
NSH-43	2.70 \pm 0.93
NSH-45	3.80 \pm 0.71
NSH-50	3.70 \pm 0.93
NSH-60	3.60 \pm 0.52
NSH-68	4.00 \pm 0.00
NSH-26 /check/	4.00 \pm 0.00

DISCUSSION

Selecting work so far in sunflower for Diaporthe resistance has been made under field conditions with an account of natural infection /Vranceanu et al. 1983; Skoric 1985/ or using artificial inoculation /Bertrand and Tourvieille 1987/. Although this type of selection has been successful and resulted in new tolerant hybrids /Skoric 1985/, the work is time-consuming and depends predominantly upon weather conditions. Yang et al. /1984/ and Sipos /1985/ indicated that stem lesions may develop of sunflower stems remaining intact at inoculation. In contrast, other investigators underlined the importance of wound-making for a successful infection /Bertrand and Tourvieille 1987; Vörös, personal communication/. We also obtained well-defined stem lesions only with seedlings that had been wounded prior to inoculation. The age of the fungal culture used as inoculum may be important as well. Yang et al. /1984/ and others considered a few-day-old mycelium as the best. In our conditions, however, agar discs colonized by 4-week-old mycelium of the fungus resulted in proper infection.

A great uncertainty may come from the subjectiveness of disease assessment. As with other lesion-producing fungi, it might be obvious to record simply the size of lesions and to calculate a disease rate from this. D. helianthi, however, causes a severe tissue necrosis within the stem, the extent of which does not necessarily corresponds to lesion size. In fact, in a number of cases we found no correlation between these two disease parameters. Instead, we concluded that the proportion of stem internal tissues showing necrosis would be a suitable sign of disease severity. In our tests, only NSH-43 exhibited good tolerance to stem canker and this result well corresponded with our previous field experience. On the other hand, NSH-45, a hybrid considered field-resistant to Diaporthe infection, did not show resistance in the growth room. It might be interesting that NSH-68 proved to be as susceptible as the control, NSH-26.

As a conclusion it is suggested that testing of sunflower for resistance to stem canker might be possible under controlled conditions where favourable environment and steady inoculation is available. In any case, the assessment should be based in part on internal tissue degeneration.

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