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Plant pathology

Effect of the scab inoculum and the susceptible parent on resistance to apple scab *(Venturia inaequalis)* in the progenies of crosses to the scab resistant cv 'Florina'

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Summary — Eight apple progenies all including the scab resistant cv 'Florina' as male parent were inoculated in the glasshouse with scab inocula originating from Angers and Wädenswil, respectively. The seedlings were inoculated twice and classified according to the reaction type. The reactions of the seedlings to the 2 inoculum sources tested were significantly different. Moreover, a clear effect of the susceptible parent on the resistance of the progeny was observed.

Malus x domestica **/ scab resistance / symptom class / inoculum source / breeding strategy**

Résumé — Effet de l'inoculum et du parent sensible sur la résistance à la tavelure due à *Venturia inaequalis* dans des descendances de croisements avec un parent résistant, le cultivar «Florina». *La création de variétés de pommes résistantes à la tavelure est très importante en vue de diminuer les interventions phytosanitaires dans les vergers. Pour que la résistance soit durable, il est indispensable de tester différents inoculums et de définir les meilleures stratégies d'amélioration génétique. Nous avons inoculé en serre, avec une suspension de conidies de tavelure, 8 descendances issues de croisements avec «Florina», porteur de la résistance «Vf», comme parent mâle. Les inoculums utilisés provenaient d'Angers et de Wädenswil. Les jeunes plantes, inoculées 2 fois et classées selon les types de symptômes, ont réagi d'une manière différente vis-à-vis des 2 sources d'inoculum. Un effet net des parents sensibles à la tavelure sur le niveau de résistance dans la descendance a été observé. Les parents les plus sensibles ont conduit à un pourcentage de plantes sensibles plus élevé dans la descendance que les parents peu sensibles. Dans les programmes d'amélioration, il est donc préférable d'associer au parent résistant des parents peu sensibles.*

Malus x domestica ***/ résistance à la tavelure / classes de symptômes / source de l'inoculum / stratégie d'amélioration***

INTRODUCTION

The development of disease-resistant apple varieties is of great importance in view of minimizing the use of chemicals in fruit production. Scab-resistant cultivars have been developed since the beginning of this century (Kellerhals, 1989). Scab caused by the fungus *Venturia inaequalis* is the most important disease in the majority of apple-growing areas. The original sources of scab resistance were small-fruited *Malus* species (Crandall, 1926). They were supposed to carry simple dominant genes (Williams and Kuç, 1969). Large fruiting material carrying polygenic scab resistance was also used (Schmidt, 1938).

Through repeated backcrossing, cultivated apple genotypes carrying resistance have been successfully developed. A range of scab-resistant varieties has already been released (Lespinasse, 1989) and many more will be commercialized in the near future. However, most of them carry the Vf-resistance derived from *Malus floribunda* 821. The nature of the Vf-resistance has still not been elucidated. Lamb and Hamilton (1969) and Rousselle *et al* (1974) postulated the presence of minor genes which can modify the level of scab resistance conferred to the Vf-resistance. Cumulative minor genes could be contributed by both the susceptible and resistant parents. According to Rousselle *et al* (1974), modifiers of major genes in *Malus* appeared to be inherited independently of major genes and to act in a quantitative manner with a mostly cumulative effect. The action of the modifiers on the major genes was identifiable in young seedlings.

An increasing number of observations question the durability of Vf-resistance (Fischer *et al,* 1983; Krüger, 1988, 1989, 1991; van der Scheer, 1989; Silbereisen, 1989). It seems that the resistance can be eroded in backcrossing if no attention is given to maintaining it through selection (Hough *et al,* 1970). Differences in scab susceptibility of old and commercial apple varieties have been documented (Aderhold, 1902; Olivier *et al,* 1984; Corbaz and Rosset, 1991; Stoll, 1991). These differences are supposed to be related to polygenes. This type of resistance could be successfully used in backcross breeding to avoid the erosion of resistance types such as Vf. We have examined the effect of the scab inoculum and of differently susceptible parents on the resistance to scab in progenies with the Vf-resistant cv 'Florina' as the male parent. In some progenies dwarf seedlings occurred. Decourtye (1967) has found a genetic link between the *Vf* gene and the recessive gene *n* for dwarfing. When dwarf seedlings occurred, the parents such as 'Florina' were heterozygous, *N/n*.

MATERIALS AND METHODS

Plant material

Eight different crosses all with the Vf-resistant cv 'Florina' as the male parent were made in 1991 at the Research Station, Wädenswil, Switzerland. The following apple cultivars were used as female parents: Vista Bella, Discovery, Alkmene, Spartan, Elstar, Arlet, Golden Delicious and Idared. Apple seeds were extracted in autumn and first stratified for 90 d at 6–8°C and then for 14 d at 0°C. At the INRA Research Center Angers, France, the seeds were sown in 73-well plastic seed trays containing a mixture of sand and compost. The progeny size varied from 88 seeds (Elstar x Florina) to 587 seeds (Arlet x Florina). The average germination rate was 84%.

Inoculation

The seedling progenies were divided into 2 equal parts. At the 3-leaf stage on the average the young seedlings were inoculated in the glasshouse with a suspension of 4.5 x 10^5 conidia/ml of *Venturia inaequalis* by using a compressor-powered sprayer. One half of each progeny was sprayed with the Wädenswil inoculum (*Venturia* strain 71, originally collected from Boscoop trees at Wädenswil without fungicide treatment and then multiplied on apple seedlings of different origin in the glasshouse). The other half of each progeny was sprayed with the Angers inoculum (collected from leaves of unsprayed Golden Delicious trees and including several scab strains from different regions in France). The conidial suspension was sprayed onto seedlings in sufficient quantities to form small droplets but without drips forming. The seedlings were incubated under plastic at 20°C and high relative humidity (rh) for 60 h. Afterwards they were kept at 20°C and 70–80% rh.

Reading of symptoms

The first reading of the symptoms was made 13 d after the inoculations using the following reaction classes (adapted from Chevalier *et al,* 1991): 0 = no visible reaction; 1 = pin-point symptom. Depression of 100–500 µm where the epidermal cells have collapsed. No subcuticular stroma; 2 = wide but shallow depressions. Limited stroma formation. No sporulation; 3a = epidermal cells collapsed over large areas. Close to the centre the abundant mycelial stromata could produce conidiophores with a limited number of conidia; 3b = lesions are a network of mycelial strands. Aborted conidiophores are mixed with normal conidiophores. Sporulating chlorosis and sporulating necrosis occur; 4 = numerous conidiophores are often grouped in clusters and sporulate abundantly. The mycelial stroma formes a dense subcuticular network.

Seedlings assigned to reaction class 4 were considered susceptible and therefore cut out; their other seedlings were marked according to their reaction classes. Immediately after the first reading a second inoculation was made with 4.7 x 10^5 conidia/ml as in the first inoculation. Fourteen d later a second reading of the symptoms was carried out. Dwarf seedlings were also considered. They were discarded later because of their poor agronomical value. Statistical analysis of the results was made using the *G*-test (Sokal and Rohlf, 1969).

RESULTS

The overall effect of the 2 scab inoculum sources on the distribution of the progenies to the different classes of symptoms following the second inoculation is illustrated in figure 1. The percentage of susceptible seedlings (class 4) was identical for both inoculum sources (56%). The ratio of susceptible (class 4): resistant (classe 0–3 b) seedlings was almost 1:1. No visible reaction (class 0) was more frequent with the Wädenswil inoculum 5.8% against 3.2% with the Angers inoculum. The number of seedlings showing the hypersensitive reaction (class 1) was generally low. Class 1 was more frequent with the Angers inoculum than with the Wädenswil inoculum. Significant differences were observed for class 2; 21.8% of the seedlings sprayed with the Wädenswil inoculum *versus* 11.9% of the seedlings sprayed with the Angers inoculum showed this reaction. In reaction class 3a the difference between the Wädenswil inoculum and the Angers inoculum was smaller.

Statistical analysis showed that the Wädenswil inoculum induced a significantly less severe reaction than the Angers inoculum. The difference was more significant after the first inoculation than after the second inoculation.

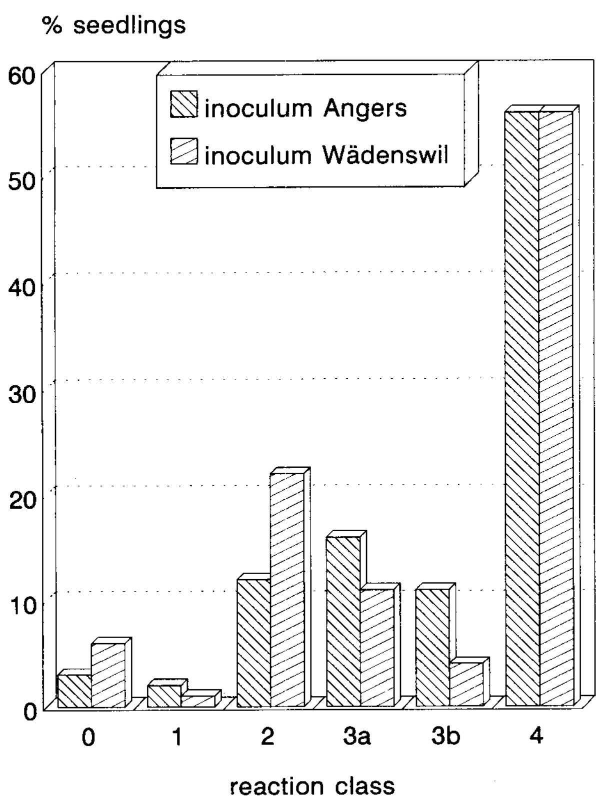

Fig 1. Effect of the scab inoculum on the distribution of the progenies to the different reaction classes after the second inoculation.

Figure 2 shows the effect of the susceptible parents on the distribution of the progeny to the different reaction classes after the second inoculation. There were statistically significant differences in the distribution between the progenies. For example, the Discovery x Florina progeny was markedly different from the Golden x Florina, Arlet x Florina and Elstar x Florina progenies for both inoculum sources. The Discovery x Florina progeny was only slightly different from the Vista Bella x Florina, Alkmene x Florina and Spartan x Florina progenies and almost the same as the Idared x Florina progeny.

In the Discovery x Florina progeny there were no individuals in classes 0 and 1, high proportions of class 2 and 3a individuals and a comparatively low proportion of class 4 individuals. The Arlet x Florina progeny consisted of a comparatively high proportion of class 0 and 1 individuals, only a few 2, 3a and 3b individuals but relatively numerous class 4 individuals. In the Arlet x Florina, the Golden x Florina and the Elstar x Florina progenies we found an average percentage of 21, 13 and 20% dwarf seedlings. The high proportion of class 0 individuals in the Arlet x Florina, Golden x Florina and Elstar x Florina progenies was due to the occurrence of dwarf seedlings which are usually found to show no reaction to the inoculation.

DISCUSSION

In this study we examined the effects of 2 different scab inoculum and 8 cross combinations on the distribution of the infection classes in the progeny. The results clearly demonstrated that the scab inoculum source had an impact on the distribution of the different symptoms of resistance in the progeny. However, following the second inoculation the difference in effect of the inocula became smaller. Finally, the percentage of susceptible (class 4) seedlings was identical for both inoculum sources. From a genetic and breeding point of view the conclusion is important: that the susceptible parent can significantly contribute to the resistance of the progeny. This result confirms the observations of Lamb and Hamilton (1969) that resistance to scab of apple progenies is influenced by the susceptible parent. In this study, we were able to demonstrate that varieties which are known to have a low susceptibility to scab such as Discovery and Alkmene, a medium susceptibility to scab such as

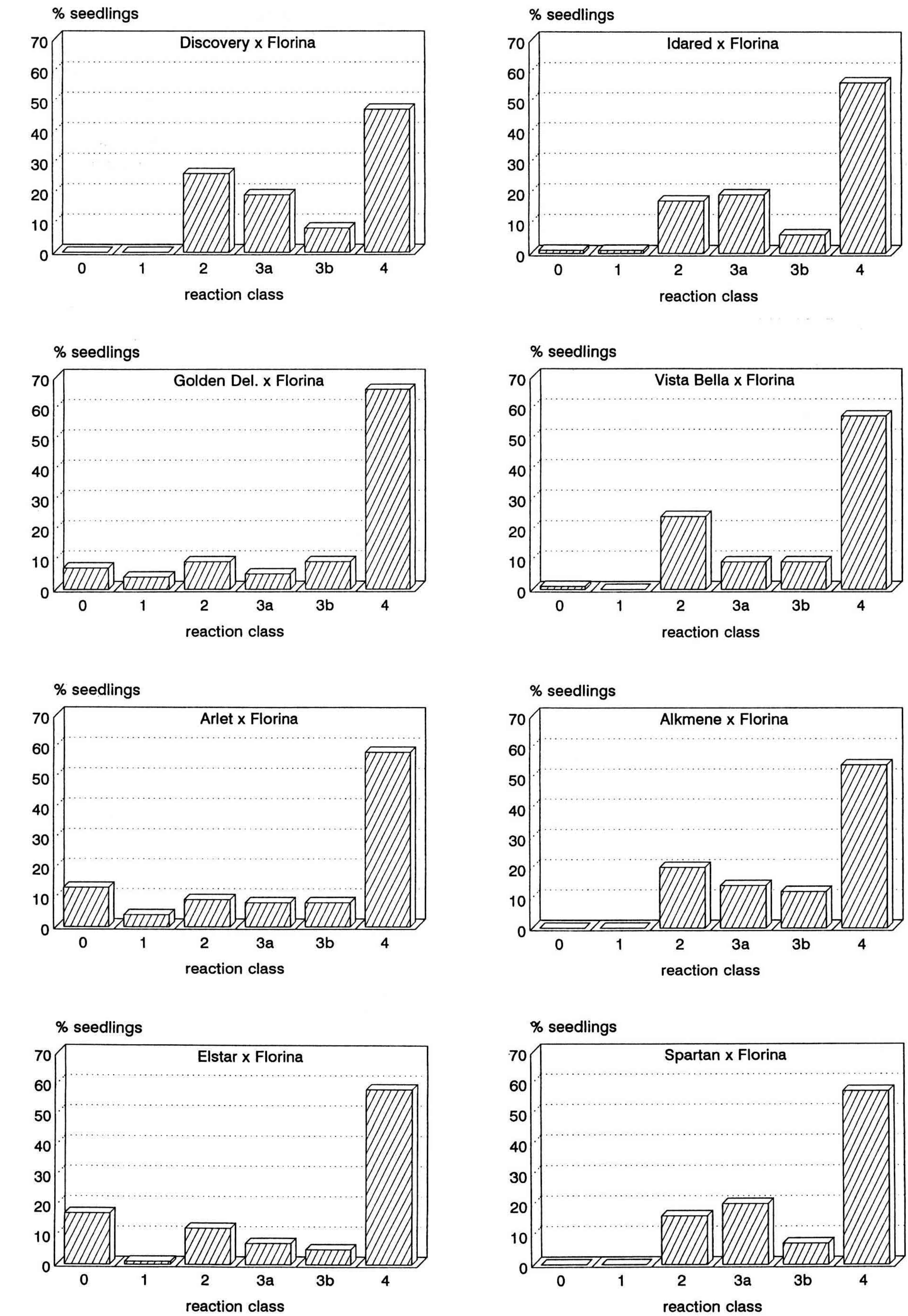

Fig 2. Effect of the susceptible parent on the distribution of the progeny to the different reaction classes after the second inoculation.

Idared, Spartan and Elstar, and a high susceptibility to scab such as Golden Delicious, Arlet and Vista Bella have an impact on the susceptibility of the progeny. Highly-susceptible parents generally induced a lower percentage of resistant seedlings in the progeny. This has already been shown by Hough (1944), Lamb and Hamilton (1969) and Rousselle *et al* (1974). However, further examination would be necessary to elucidate whether resistant individuals in a progeny originating from a not very susceptible parent are able to transmit a stronger resistance than similar individuals originating from a progeny with a highly susceptible parent.

Dwarf seedlings were found in 3 progenies at a percentage close to 25%. They mostly did not show any reaction to the infection. It is obvious that their leaf structure is different from that of normal seedlings. It would be interesting to compare the histological structure of leaves from normal and dwarf seedlings and to compare the penetration of the scab fungus.

The results of our experiments show that combining parents carrying a polygenically determined low susceptibility to scab with parents carrying Vf-resistance or an other mono- or oligogenic resistance might offer interesting prospects in view of strengthening the resistance. One reason for the erosion of resistance through repeated backcrossing might be the choice of very scab-susceptible parents.

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