An Assessment of the Durability and Susceptibility of Scab Resistance in Apple Cultivars

H. Kemp[1], M.P. van der Maas[1], R.E. Voorrips[2], R. Groenwold[2] and W.E. van de Weg[2]
[1]Applied Plant Research sector Fruit P.O. Box 200, 6670 AE Zetten, The Netherlands
[2]Plant Research International, P.O. Box 16, 6700 AA Wageningen, The Netherlands

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Abstract

Durable resistance to scab (*Venturia inaequalis*) of newly introduced cultivars is needed to achieve a considerable reduction of pesticide use. However, the assessment in the field on the level and durability of scab resistance is time and space consuming. A proper and fast indoor test should be a convenient tool to discard cultivars with a limited level of resistance at an early stage. As a result, field tests can include only the better cultivars. This approach has been tried out by comparing field data with data from a glasshouse test, comprising twelve new and seven well-known susceptible and resistant (with and without *Vf*) cultivars which have been tested with six isolates of scab from different origins, virulence and aggressiveness, including race 6 and 7. Young trees on M.9 rootstock were infected and after a 17 day period at high humidity, sporulation and chlorosis have been observed. If the set of isolates is broad and well-chosen it seems possible to assess susceptibility and possibly also durability. To optimize the choice of isolates, it is desirable to know the pedigree of the cultivars that are tested. In addition, this procedure allows the use of highly virulent and aggressive races, that should be avoided in field tests for the risk of spreading.

INTRODUCTION

In areas with a high risk of scab infection, resistant cultivars are useful to achieve a reduction of pesticide use. For exploitation by growers, resistance has to be durable. In order to make predictions on the durability of scab resistance prior to introduction or large scale planting, 12 new cultivars have been tested in a glasshouse with a wide range of isolates, together with 7 well-known reference cultivars. Assessment in the field on the level and durability of scab resistance is time and space consuming. If proven appropriate, a glasshouse test should be a convenient alternative.

MATERIALS AND METHODS

Seven reference cultivars (‘Gala’ and ‘Golden Delicious’ as highly susceptible, ‘Santana’ and ‘Topaz’ as *Vf*-resistant and ‘Discovery’, ‘Alkmene’, and ‘Durello di Forli’ as low susceptible) and 12 new cultivars were chip budded on M.9 rootstock in August 2001. They were grubbed in winter and stored in air (0-1°C) till approximately 5 weeks before inoculation. Trees were raised at 16°C. Six isolates – among which four characterised in the DARE project (Parisi et al., 2004) – were used (Table 1), which differ in origin, virulence and aggressiveness and include race 6 and 7 which overcome the *Vf*-resistance (Bénaouf and Parisi, 2000). The PPO-isolate was a mass-isolate obtained from mixed leaves of ‘Elstar’, ‘Golden Delicious’, and ‘Gala’ grown in an almost fungicide free cultivar trial plot at Randwijk.

Each cultivar/isolate combination was tested on 5 plants. All isolates were applied with 5.10^5 spores/ml. After inoculation the plants were kept in darkness for 40 hours at 18°C, 100% RH and afterwards grown for 17 days in a glasshouse at 18°C and 80-90% RH. As a reference for normal plant development, one plant was “inoculated” with water only. After 17 days, sporulation and chlorosis were observed on the first seven fully unfolded leaves, starting from the top and from the moment of inoculation: sporulation as a % of leaf surface (0 = no symptoms, 1 = 0-1, 2 = 1-5, 3 = 5-10, 4 = 10-25, 5 = 25-50, 6 = 50-75, 7 >75% (Croxall et al. (1952) modified by Parisi et al., 1993) and chlorosis as a

% of yellow leaf area (0 = no symptoms, 1 = 0-25, 2 = 25-50, 3 = 50-75, 4 > 75%). The Chevalier scale (Chevalier et al., 1991) was not used because chlorosis mostly occurred in the case of no sporulation. Practically all these leaves would have had Chevalier-score 2, which would not add information.

For statistical analysis, the scores were transformed to fraction of leaf surface. To enable ANOVA and REML analyses, a second transformation was done. Data given in Table 1 and 2 were obtained after retransformation to leaf fraction. The data were compared with results (not given) obtained in an almost unsprayed test orchard, from which the PPO mass-isolate was obtained.

RESULTS

Table 1 gives the sporulation data on leaves 1-7. Genotypes and isolates showed highly significant interactions, indicating that a considerable part of the observed resistances is race specific, and therefore not durable. ‘Discovery’, ‘Durello di Forli’, and 0062 were resistant to all isolates, while ‘Gala’ and 5 new cultivars were susceptible. ‘Santana’, ‘Alkmene’, 9750, and 99127 were rather resistant to five isolates. The resistance of ‘Topaz’ and ‘Golden Delicious’ and 3 cultivars was proved to vary per isolate. ‘Topaz’ is highly susceptible for three *Vf* compatible isolates, while ‘Golden Delicious’ was resistant to just one isolate (1066). This latter resistance is known to be due to the resistance gene *Vg* (Durel et al., 2000), which is also present in ‘Discovery’ (Calenge et al., 2004).

The current conclusions are based on the average sporulation area of leaves 1 to 7. However, these conclusions were also identical when only leaves 2 and 3 would have been considered. So, in future, fewer observations suffice. In general, leaves 2 and 3 showed also the highest level of sporulation.

In addition to sporulation, chlorosis was also assessed. These parameters were not correlated with regard to area, but they were for leaf position: the leaf with the largest fraction of sporulation also showed the largest fraction of chlorosis. So, older leaves also showed less chlorosis. Chlorosis was less isolate specific than sporulation. No relation was found between average sporulation and average chlorosis per genotype. Only the chlorotic leaf fraction of the non-sporulating leaves 2 and 3 is given in Table 2.

CONCLUSIONS AND DISCUSSION

A glasshouse test with a broad and well-chosen set of isolates seems effective to quickly identify cultivars that have resistance to only a limited number of fungal isolates. This makes subsequent field trials more effective and cost-efficient since now only cultivars can be included that are likely to have resistance to a wide series of races. The task remaining is to distinguish the very best from the best. It proved very useful to include aggressive or highly virulent isolates in glasshouse tests, of which testing in the field is not desired for the risk of spreading.

The data indicate that our current set of isolates have to be further optimised. Two cultivars were severely diseased in the experimental plots, while the glasshouse test indicates the presence of a high level of (incomplete) resistance to some or all tested isolates. This divergence indicates that our current series of isolates is not yet broad enough to cover the current genetic variation of the fungus, as was expected. With the genotypes KA 0105, 99029, 0060, and 9614 that proved to be susceptible both indoors and in the field (data not given), the series of isolates was broad enough for a proper assessment, as it was with ‘Golden Delicious’ and ‘Gala’.

The power of our approach is illustrated by ‘Topaz’. To date ‘Topaz’ has not been infected in the experimental field at PPO in Randwijk. However, the current data clearly show this cultivar to be highly susceptible to three *Vf* compatible isolates. It indicates that the natural scab population in our experimental field is short of *Vf* compatible races. In this field we apply 1-4 fungicide applications during the risky ascospore period to prevent breakdown of *Vf*. For fruit growers this is essential too, to minimize the risk of breakdown, especially with ‘Topaz’, according to our results. As with ‘Topaz’, we found with the supposed “polygenic” genotype 99069, to have a low infection in the field, but high in the indoor test.

According to Bénaouf and Parisi (2000), ‘Golden Delicious’ is completely resistant to isolate 1066. We also found a relatively low infection by 1066, but the score was significantly higher than 0, which result is consistent with that of Durel et al. (2000).

In addition to supporting the methodology, this research gave valuable information on the level of resistance of some new cultivars. The best new selection was 0062, which was highly resistant to all isolates, like the ‘polygenic’, durably resistant cultivars ‘Discovery’ and ‘Durello di Forli’. Selection 0062 also showed low levels of chlorosis, while ‘Durello di Forli’ and ‘Alkmene’ showed quite a lot. This new selection is known to carry *Vf*, however, few scab symptoms were found in the field. Selection 99127 was also interesting, because of its low levels of sporulation and chlorosis. It seems to carry *Vf* and possibly *Vg*. Selection 0222 is supposed to have V*f*, which is confirmed by our data. Cultivar 0030 reacted the same as ‘Alkmene’ and ‘Durello di Forli’: outdoor and indoor a low level of sporulation but with quite a lot of chlorosis.

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Tables

Table 1. Average leaf area fraction with sporulation of leaves 1-7 investigated in glasshouse.

Genotype	Isolate [1)] NL19	NL61	PPO	EU-D-42	1066	NL24	Avg.
0182	0.11	0.00	0.06	0.34	0.10	0.00	**0.07**
KA 0105	0.37	0.38	0.61	0.21	0.40	0.39	**0.39**
99069	0.25	0.41	0.24	0.11	0.24	0.10	**0.22**
99029	0.34	0.33	0.51	0.35	0.38	0.52	**0.41**
0060	0.02	0.05	0.28	0.14	0.33	0.34	**0.17**
0062	0.01	0.00	0.00	0.00	0.00	0.00	**0.00**
9614	0.10	0.14	0.31	0.56	0.38	0.28	**0.28**
99127	0.00	0.00	0.00	0.16	0.06	0.24	**0.04**
0030	0.06	0.00	0.02	0.04	0.03	0.16	**0.04**
9750	0.09	0.12	0.07	0.09	0.07	0.06	**0.08**
Santana	0.00	0.00	0.00	0.08	0.06	0.22	**0.03**
Discovery	0.00	0.00	0.00	0.00	0.00	0.00	**0.00**
Alkmene	0.00	0.01	0.01	0.04	0.04	0.13	**0.03**
Durello di Forli	0.00	0.00	0.00	0.00	0.03	0.00	**0.00**
Golden Delicious	0.35	0.39	0.61	0.39	0.04	0.37	**0.34**
Topaz	0.00	0.00	0.02	0.74	0.44	0.19	**0.15**
0222	0.00	0.00	0.08	0.10	0.03	0.13	**0.04**
Gala	0.19	0.20	0.44	0.35	0.27	0.38	**0.30**
Avg.	**0.07**	**0.06**	**0.12**	**0.17**	**0.13**	**0.17**	**0.11**

[1)] EU-numbers are isolate codes from the EU DARE project, NL19 = EU-NL-19, NL24 = EU-NL-24, and NL61 = EU-NL-37. EU-D-42 previously known as race 6 and 1066 as race 7.

Table 2. Average leaf area fraction with chlorosis of leaves 2 and 3 of non-sporulating race:genotype combinations, investigated in glasshouse.

Genotype	Isolate [1)] NL19	NL61	PPO	EU-D-42	1066	NL24	Avg.
0182	*	0.45	0.87	*	*	0.56	**0.64**
KA 0105	*	*	*	*	*	*	*****
99069	*	*	*	*	*	*	*****
99029	*	*	*	*	*	*	*****
0060	0.34	0.87	*	0.71	*	*	**0.66**
0062	0.00	0.22	0.00	0.06	0.00	0.00	**0.02**
9614	*	*	*	*	*	*	*****
99127	0.00	0.01	0.00	*	0.00	*	**0.00**
0030	0.87	0.26	0.87	0.80	0.87	*	**0.76**
9750	*	*	*	*	*	*	*****
Santana	0.03	0.01	0.05	*	*	*	**0.02**
Discovery	0.03	0.26	0.16	0.19	0.12	0.29	**0.16**
Alkmene	0.64	0.82	0.87	0.73	0.87	*	**0.80**
Durello di Forli	0.53	0.72	0.87	0.37	0.87	0.42	**0.65**
Golden Delicious	*	*	*	*	*	*	*****
Topaz	0.64	0.20	*	*	*	*	**0.41**
0222	0.00	0.00	*	*	*	*	**0.00**
Gala	*	*	*	*	*	*	*****
Avg.	**0.22**	**0.29**	**0.40**	**0.46**	**0.38**	**0.26**	*****

[1)] see Table 1

* None or almost no non-sporulating leaves present