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# Strawberry breeding for disease resistance in Dresden

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# Abstract

Verticillium resistance is one of the most important breeding goals in strawberry resistance breeding at Dresden-Pillnitz. Resistance evaluation of cultivars, advanced selections and seedlings is realized under natural conditions at a provocation field and by artificial inoculation in the greenhouse. Introgression of Fragaria chiloensis L. (Miller) into Fragaria xananassa Duch. resulted in highly tolerant breeding selections. After back-crossing with cultivars of F. xananassa first genotypes were selected which can be evaluated in experimental cultivar trials at different locations in Germany.

Keywords: strawberry breeding, disease resistance, Fragaria, Verticillium

### Introduction

Breeding in strawberry was started at Dresden-Pillnitz, Germany in the 1920s when new cultivars were developed for home gardening. Since that time the aims of selection as well as the methodology has been changed continuously according to the demands of producers, fresh market, processing industry, and consumers. Integrated and biological methods of fruit production became increasingly important. Strawberry breeding is focused on two main problems: (1) increased disease resistance to overcome restrictions in plant protection and to minimize the application of pesticides, and (2) fruit quality to improve attractiveness, firmness and aroma composition of the fruit.

Beside the bacterial disease caused by *Xanthomonas fragaria* Kennedy & King, fungal diseases are most important in strawberry production. Since the 1980s resistance to soil born diseases, especially *Verticillium* wilt (*Verticillium albo-atrum* Reinke & Berthold and *V. dahliae* Kleb.) has been one of the main topics in breeding programs at Dresden-Pillnitz (Rössel & Büttner 1989; Olbricht *et al.*, 2006). The evaluation of plant material for resistance to *Verticillium* is performed in the greenhouse using an artificial inoculation procedure as well as in provocation fields under natural conditions. The methodology is applied for characterization of seedlings, advanced selections and cultivars of *F. xananassa* but also wild species of the genus *Fragaria*.

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# **Material and Methods**

### Plant material

Cultivars of *F.* ×ananassa as well as advanced selections resulting from cross breeding with *Fragaria chiloensis* subsp. *lucida* (E. Vilmorin ex Gay) Staudt were evaluated for resistance (Tab.1). The cultivars Fraroma and Senga Sengana were used as standard types for high tolerance, Elsanta and Honeoye as susceptible cultivars. In each resistance trial under greenhouse conditions, 10 plants per genotype were inoculated, five plants each served as control. In the field trial, blocks of 18 plants were evaluated.

### **Resistance Evaluation**

A suspension of at least ten isolates from different German provenances containing conidia and microsclerotia was used for resistance evaluation in greenhouse. A concentration of  $10^7$  conidia per ml was used. Microsclerotia were not counted for the suspension. Roots were cleaned from soil by water; one third of the root length was cut off. For artificial inoculation, the roots of plants were placed into the suspension (control plants in water) for 24 hours before plants were potted and cultivated in greenhouse as described by Olbricht *et al.* (2006).

The greenhouse evaluation was performed in the years 2004 to 2007 with at least one replication. The evaluation was performed based on the definition of five degrees for disease severity in the strawberry plant:

- 1 no symptoms on plants
- 3 wilting symptoms on single outer leaves
- 5 wilting symptoms on all outer leaves
- 7 all leaves are wilting except leaves in the heart of the plant
- 9 plants killed

For field evaluation under natural conditions a provocation field was used which is highly infested with *V. dahliae*. The evaluation was performed in the years 2003 to 2007 with at least one replication.

Re-isolation of the *Verticillium* fungus from petioles of inoculated plants was performed in order to describe whether the fungus invaded the plant (Olbricht 2004).

# Results

Seedling populations of cross-breeding with *F. chiloensis* subsp. *lucida* were characterized by a high degree of resistance to *Verticillium* based on artificial infection. None of the seedlings died or showed any wilting symptoms under natural conditions of infection in the field. Additionally the glossy leaf surface, typical for this subspecies (Staudt, 1999), is strongly inherited in the F1 generation and apparently protects the plant from infection by other fungi causing diseases in leaves, such as powdery mildew, leaf scorch, and leaf spot (Olbricht et al. 2006).

Two selections 97/362 and 97/369 were selected from populations where Elsanta respectively Honeoye were used as mother cultivars and *F. chiloensis* subsp. *lucida* as father. Results obtained in resistance evaluation both in greenhouse and in the field were confirmed so far (Tab. 1). Re-isolations of the fungus from petioles of the selections 97/362, 97/369 and of the highly tolerant standard cultivars Fraroma and Senga Sengana showed that plants can survive with the fungus inside without having wilting symptoms. However, a depressed plant growth was detected in comparison to control plants (Rössel & Büttner, 1989).

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Back-crossing of 97/362 and 97/369 with different cultivars of *F.* ×ananassa resulted in vigorous and breeding clones, without wilting symptoms at the provocation field and fruits developed an extraordinary flavor. The important key compound of flavor, methyl anthranilat (Ulrich *et al.*, 1997; Olbricht *et al.*, 2007), could be maintained up the F2 generation so far (unpublished data). Two advanced selections, P-5357 and P-5284 resulting from introgression of *F. chiloensis* subsp. *lucida*, are announced for experimental field trials at different sites in Germany. Beside good taste, sufficient firmness and yield, they might be of interest especially for organic fruit production due to their high tolerance to *Verticillium* wilt and other fungal diseases.

Cultivar/ Selection	Resistance evaluation	
	Severity in the field	Severity in the greenhouse
Elsanta	3,5,7,9	9
Honeoye	3,5,7,9	9
Fraroma	1	1
Senga Sengana	1	1
Florence	1	3,5
Susy	1,3	3,5
Polka	1,3	3,5
Korona	1,3,5	3,5
Thutop	3,5,7,9	7,9
Nancy	3,5	3,5
97/362	1	1
97/369	1	1
P-5357	1	not tested
P-5284	1	not tested

Table 1: Resistance to Verticillium dahliae under field and greenhouse conditions

Severity: <u>highly tolerant</u>: 1-no symptoms on plants; <u>medium susceptible</u>: 3-wilting symptoms on single outer leaves; 5-wilting symptoms on all outer leaves; <u>susceptible</u>: 7-all leaves are wilting except leaves in the heart of the plant; <u>highly susceptible</u>: 9- plants killed

# Discussion

*Verticillium* is a serious fungal disease in strawberry. *Verticillium* species are root invaders. They do not live in the soil as saprophytic fungi but can survive in the soil for several years as microsclerotia. Infected dead root systems improve the survival in the soil. The *Verticillium* fungi are often moved with infected soil or plants. The plant symptoms that result when this fungus attacks may be confused as seasonal die-back of foliage and may occur together with other plant problems such as fusarium wilt, bacterial wilt, root rots as well as drought and damage due to excessive soil moisture. Plants may be infected for a while before symptoms become visible. Initially plants wilt. Witting tissue soon begins to yellow then turn brown and die. Planting of tolerant strawberry cultivars is the most important and effective measurement to minimize the beds from being infested with *Verticillium* wilt.

We presented results on breeding for resistance to *Verticillium*. The presented selections resulting from introgression of *F. chiloensis* subsp. *lucida* and back-crossing with cultivars of *F. ×ananassa* show the possibility to obtain valuable material already in a F2 generation which is characterized by a high level of tolerance to the pathogen. It should be also mentioned that the term "resistance" used for *Verticillium* wilt throughout the literature is not very precise (Newton & van Adrichem, 1958; Tahmatsidou *et al.* 2002). Only few authors defined *Verticillium* resistance of strawberry cultivars, breeding selections or species in the strict sense of its meaning. As the pathogen can be isolated from plants not showing wilting symptoms, this is an indication for high tolerance but not for a resistance reaction. (Zinkernagel, 1970; Rössel & Büttner, 1989; Olbricht 2004; Olbricht *et al.*, 2006). On the other hand, there are cultivars susceptible to *Verticillium* after inoculation in the greenhouse but showing a high field resistance under natural conditions, for example the cultivar Florence.

In order to supply material for test trials at different locations throughout Germany, micropropagation of the new selections starting from meristem cultures was initiated. That material will be available in 2009 for field trials. Following a new system of handling the propagation rights, the cultivars will be available for all kinds of cultivation including organic production.

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