## Use of genetic resources and partial resistances for apple breeding

Nutzen von Genressourcen und Teilresistenzen in der Apfelzüchtung

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### **Abstract**

Modern apple breeding strategies are mainly considering the most advanced selections and cultivars as parents. This tends to lead to a narrowed genetic basis. The introgression of traditional varieties and accessions of the gene pool is often feared due to undesirable characteristics that might be incorporated. However, there is scope for considering a wider genetic basis in apple breeding to support sustainable fruit production systems. The focus at Agroscope Changins-Wädenswil (ACW) is put on high fruit quality combined with low susceptibility to scab (Venturia inaequalis), fire blight (Erwinia amylovora), and powdery mildew (Podosphaera leucotricha). The Swiss scab and fire blight resistance research programs are focused on developing and applying molecular markers for resistance factors. In this context we are also mining our national germplasm collections of heritage varieties. Heritage varieties are traditional and/or special use apple varieties. Many of them were discovered in the course of a national inventory over the past 5 years. As a result of this inventory, 1'100 apple and 670 pear accession are being added into the germplasm collections. They will be evaluated in the coming years for different fruit and tree characters.

Keywords: apple genetic resources, scab, fire blight, resistance

#### Introduction

Durable disease resistance is one of the main objectives in apple breeding worldwide. Host resistance is a safe, reliable, and low input option that might be a key component of integrated disease control strategies. Such resistant cultivars grown in adapted and sustainable systems could further improve the image of apples being a healthy food and contribute to high food safety. Scab (caused by the fungus *Venturia inaequalis*), fire blight (caused by the bacterium *Erwinia amylovora*), and powdery mildew (caused by the fungus *Podosphaera leucotricha*) are the major diseases of apple. The fungal diseases affect apple quality and yield when they are not strictly controlled, most commonly with repeated fungicide applications. In organic and integrated production systems this control is often difficult. Apple breeding strategies, incorporating the resistance based on single genetic factors from related wild species, such as *Vf, Vr*, etc. conferring scab resistance, have been applied to develop new resistant varieties for many decades. However, most of these genetic factors can with time be overcome by new pathogen races. New ways of breeding are required in order to achieve durable resistance in apple. Partial, often called 'polygenic' resistance should give more durable resistance, especially if combined with 'monogenic' resistance.

Current options in Europe for controlling the most serious bacterial disease of apple, fire blight, are mainly limited to prevention and sanitation measures with some progress in developing biocontrol. There is scope to develop new varieties with low susceptibility to fire blight.

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#### **Material and Methods**

At ACW we are selecting progenies for their fruit quality which have been bred from partially resistant parents and/or parents that do not carry a major gene resistance against scab such as *Vf.* The progenies originate from di-allele crosses which were made in 1997 and 1998 including the following cultivars and selections: 'Zitronenreinette', 'Oberrieder Glanzreinette', 'Discovery', 'Delbard Jubilé', 'Champagner Reinette', 'Pilot', 'Angold', 'Reglindis', A 810-390 (Pl2), 'Schweizer Orangenapfel', HL 75-27 ('Goldspur' x 'Antonovka'), HL 10-04 ('Goldspur' x 'Glockenapfel') and 'Nabella'. The first selections are being multiplied for advanced testing e.g. FAW 13961 ('Zitronenreinette' x 'Discovery'). Later crosses were established and screened in similar ways. Results from the following crosses made in 2002 are shown:

- Delrouval x FAW 12309
- FAW 5878 x Rubinstep
- La Flamboyante x FAW 5878

The parentage of these varieties is given in Table 1.

Table 1: Parentage of varieties used in crosses to achieve selections with partial disease resistance

Variety	Mother	Father	
Delrouval	Delcorf	Primerouge (Akane)	
FAW 12309	Discovery	Delbard Jubilé	
FAW 5878	(Idared x Maigold)	Elstar	
Rubinstep	Clivia	Rubin	
La Flamboyante	Gala	Maigold	

The selection for scab resistance is usually performed on the young seedlings in the glasshouse with a high dosage inoculation of 350'000 scab conidia per ml under controlled temperature and humidity. Scab symptoms are evaluated about two weeks after inoculation according to the scale of Chevalier et al. (1991). For progenies, where partially resistant parents were used, about 10 times lower inoculation dosages are applied. In the past, we also used the scale of Chevalier et al. (1991) to select partially resistant plants in these progenies. Two years ago we have switched to a new quantitative symptom evaluation scale developed by Lefrancq et al. (2004) especially for progenies incorporating partial scab resistance. After the glasshouse test the seedlings are planted in a field nursery where no fungicides are applied. Scab and mildew incidences are scored in the second year using the scale adapted from Lateur and Populer (1994) where 1 = no incidence and 9 = tree completely infected.

A national inventory of major and minor fruit genetic resources in Switzerland started in 2000 and was completed in spring 2005. The project was financed by the Ministry of Agriculture and conducted by the NGO Fructus at ACW (Kellerhals and Egger, 2004). It is the basis for a complete and safe conservation of fruit genetic resources in Switzerland. In the course of the inventory, a fundamental project of the national plan of action, plenty of information was collected related to the origin, abundance and frequency of accessions. We have started to screen this gene pool for partial resistances against scab, fire blight and mildew.

The screening of heritage varieties for relative fire blight tolerance was conducted in the quarantine glasshouse at ACH in Wädenswil. Scion material was grafted onto M9 derivative rootstock and overwintered under refrigeration. In the spring, trees were planted in plastic deep-pots 60 from Stuewe & Sons (Corvallis, US) with a length of 35.5 cm and diameter of 7 cm and then grown in the glasshouse for several weeks prior to inoculation. For each variety, 8-10 replicate trees were inoculated by puncturing the distal tip of shoots 15-30 cm long with a syringe containing an *E. amylovora* solution of 10<sup>6</sup> cfu/ml. Disease was evaluated weekly for four weeks by measuring the expansion of

necrotic lesion from the tip in relation to the total shoot length. Artificial fire blight infections were performed in 2005 with 23 different accessions, including heritage varieties still grown for apple juice and cider production, and with commercial standard varieties.

#### Results

## Partial scab resistance

Table 2 shows examples of crosses including partial scab and mildew resistance. In the glasshouse test we discarded more than 60 % of the progeny seedlings of 'Delrouval' x 'FAW 12309' and 'La Flamboyante' x 'FAW 5878', but only 14% in the 'FAW 5878' x 'Rubin' progeny applying the scale of Chevalier et al. (1991). In progenies carrying 'monogenic' scab resistance, after discarding the susceptible plants following the glasshouse test, there are usually no scab susceptible seedlings observed in the field. However, when working with partial resistance, the situation is different (Table 2). While scoring the scab and mildew incidence on the two year old seedlings in the field, the 'Delrouval' x 'FAW 12309' population was the healthiest. These plants were more vigorous mainly because they were less damaged by scab and mildew. 'FAW 5878' x 'Rubinstep' was less tolerant. Mildew infection was rather high. The lowest tolerance was found in the combination 'La Flamboyante' x 'FAW 5878' as could be expected from the parents. Mildew and scab incidence were strong in this progeny.

**Table 2:** Selection for partial scab resistance in the apple breeding program at ACW in Wädenswil

Cross	No. of Seed-	Seedlings in	No. seedlings	Scab incidence	Mildew inci-	No. of seed-
	lings	scab class 4* in	planted to the	on the seed-	dence on the	lings selected
		the glasshouse	field (%)	lings in the field		for fruiting
		test		in 2 <sup>nd</sup> leaf	field in 2 <sup>nd</sup> leaf	stage
				(2004)**	(2004)**	
Delrouval x	968	585 (60.4%)	383	2.74	5.09	40
FAW 12309						
FAW 5878 x	590	84 (14.2%)	506	1.68	6.78	38
Rubinstep		,				
La Flamboy-	1413	934 (66.1%)	479	4.09	7.76	17
ante x FAW		, , ,				
5878						

<sup>\*</sup> according to Chevalier et al. (1991)

## Fire blight testing

The preliminary results of fire blight testing in the glasshouse revealed a wide range of tolerance and susceptibility among the apple accessions (Figure 1). Continuing tests will further evaluate accessions of important interest from the Swiss fruit genetic resources. Our results are in general agreement with those previously reported in Europe and North America for most varieties. Most varieties displayed no resistance and were highly susceptible. However, a few varieties held back the advance of the pathogen. 'Spartan' was highly resistant in our trials and will be retested in 2006. Information such as this, and when added to similar data from trials elsewhere, on the susceptibility will enable growers that express an interest in preserving and exploiting heritage variety plantings (eg. in cider apple orchards, in dairy pastures for shade and landscape value) to make informed choices as to which varieties to replant and which to avoid due to fire blight sensitivity.

<sup>\*\*</sup> according to Lateur and Populer (1994)

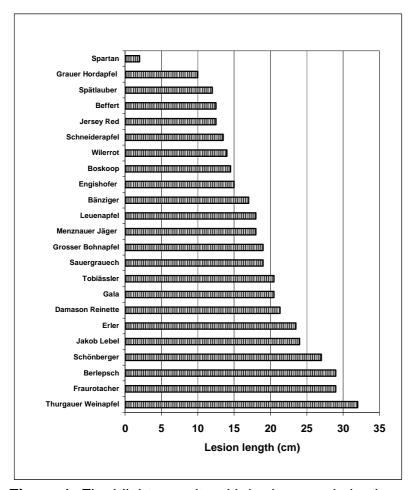


Figure 1: Fire blight severity with heritage varieties in a glasshouse infection test

## **Discussion**

There is scope for breeding fire blight resistant apple and pear cultivars by exploiting genetic variation in germplasm and by developing QTL markers. In a preliminary glasshouse screening test with apple germplasm we have detected considerable differences in susceptibility. Peil et al. (2004) analyzed fire blight infections resulting from a natural outbreak in the genebank of the Institute of Fruit Breeding in Dresden-Pillnitz in 2003. They found a correlation between the susceptibility in the field and after artificial shoot inoculation with fire blight in the glasshouse for the resistant varieties (Reseries from Dresden) only and not for the conventional varieties (Pi-series). Rewena showed an especially good resistance level.

Forsline and Aldwickle (2002) have screened the USDA Apple Collection at Geneva N.Y. including apple germplasm from Asia and Europe for natural occurrence of fire blight. For fire blight no major resistance genes have been found. Therefore it seems reasonable to opt for QTL's. Dondini et al. (2004) have found QTLs linked to fire blight in pear and Calenge et al. (2005) found a major additive QTL for fire blight resistance based on work with two related apple progenies, and we have been able to confirm this QTL in apple at ACW in Wädenswil (A. Khan, B. Duffy, A. Patocchi, unpublished data). The use of genetic resources and partial disease resistance are promising for the development of diverse sustainable apple production systems with low pesticide input. As MacHardy et al. (2001) stated, all major resistances in apple such as *Vf, Vm, Vr* are ephemeral. It is therefore necessary to define alternative strategies for durable resistance. Besides the pyramiding of major

genes the inclusion of partial resistance present in genetic resources, breeding material, and some commercial varieties might be promising. In the framework of the European DARE project local European cultivars were examined as sources for durable scab resistance in apple (Laurens et al. 2004). During this research very diverse and complex resistance behaviors have been found. For example, the cultivars which showed the widest range of resistance were mostly local cultivars and some newly selected hybrids combining major genes and partial resistance. Inventorying, collecting, characterizing, evaluating and utilizing the fruit genetic resources, as in a breeding program, are of great public relevance. It allows keeping and utilizing a heritage for future generations, to broaden the genetic basis in breeding programs and to meet consumers evolving demands of healthy and new and innovative products.

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