

## USE OF GENETIC RESOURCES IN APPLE BREEDING AND FOR SUSTAINABLE FRUIT PRODUCTION

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### A B S T R A C T

The public awareness about the worldwide loss of genetic diversity in cultivated plant species has considerably increased in recent years. This concern has fostered support for genetic resources conservation. It has been recognized that conserving the genetic diversity in fruit species maintains a relevant and not substitutable access to a source of fruit quality, disease and pest resistances and growing types useful for breeding. In this paper the achievements and the potential of genetic resources in modern breeding in favour of sustainable apple production, are analysed.

**Key words:** apple breeding, disease resistance, genetic resources, sustainability

### INTRODUCTION

The United Nations Conference on Environment and Development was held in Rio de Janeiro in 1992. A key agreement adopted was the Convention on Biological Diversity (CBD). The convention has fostered international activities for the conservation and sustainable use of plant genetic resources for food and agriculture. It establishes three main goals: the conservation of biological diversity, its sustainable use, and the sharing of benefits from their use. As in many crops, also in fruit species the genetic diversity has considerably decreased in the last decades. A few world wide grown fruit species and cultivars became predominant. In Switzerland, based on the global plan, a national plan of action for the conservation and sustainable use of plant genetic resources for food and agriculture was adopted. Subsequently crop related concepts were developed and projects supported that deal with

the objectives of the plan of action. The steps include an inventory of the resources still available, their conservation, characterization and use.

Breeding strategies are most often considering the most advanced selections and cultivars and thus leading to a narrow 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 for sustainable fruit production.

### Breeding aims

Modern breeding aims in apple (*Malus x domestica* Borkh.) are determined by market and consumer requirements. The markets become more and more international. This leads to the cultivation of world apples such as ‘Golden Delicious’, ‘Gala’, ‘Fuji’ and ‘Braeburn’. Besides, additional cultivars that are locally produced and desired by consumers especially in periurban growing zones, are available. As part of sustainable systems, cultivars need to satisfy economical and ecological demands. Since market mandate, new and successful cultivars must be superior in some specific traits than the presently marketed ones. This means that they must be distinctive and improved with respect to intrinsic and extrinsic quality aspects. For addressing ecological concerns, there is an intensive effort to develop disease resistant high quality cultivars. Disease resistant varieties allow a significant reduction of pesticide inputs in orchards in temperate climates where compared to other crops more treatments are needed (e.g. Fried et al., 1992). Such cultivars grown in adapted and sustainable systems could further improve the image of apples being a healthy food and contribute to high food safety. However, none of those disease resistant cultivars has yet achieved a breakthrough in the marketplace comparable to varieties such as ‘Golden Delicious’, ‘Gala’ or ‘Braeburn’. The success of disease resistant varieties at the point of sale is not related to their attribute ‘disease resistant’. The successful introduction of a new cultivar is primarily determined by wholesalers and retailers, and these refer to consumer needs, as they perceive them, relating to fruit quality and a respective marketing strategy. A recent consumer test performed by Agroscope FAW Wädenswil in a local supermarket revealed fruit quality criteria to be more important to consumers than the attribute ‘from organic production’ (Tab. 1).

Table 1. Importance of different quality and production criteria to consumers in a survey in Switzerland, June 2004, 200 consumers, in percent

	Firmness	Crispness	Flavour	Organic production
Not important	10	6	5	57
Important	51	43	47	25
Very important	39	51	48	18

Moreover, on the production side, disease resistant cultivars need to be durable in respect to resistance attributes. The breakdown of resistances as observed in some geographical areas, especially in the northern part of Europe, with apple cultivars carrying the *Vf* scab resistance has disillusioned those who thought, that with a respective cultivar, the key disease and pest problems could be resolved. However, we know from other pathosystems, that resistance genes may be of durable use if properly managed and not “left alone”, to resolve pressing crop protection problems†.

### **Recurrent breeding strategies**

The Horticulture and Food Research Institute of New Zealand Ltd (Hort Research) has established an Apple Genetics Population to maintain biodiversity for cultivar development (Noiton and Alspach, 1996) and to provide genetic information on important apple characteristics (Oraguzie et al., 2000; 2001). The population is anticipated to provide novel fruit characteristics for application in a long-term breeding strategy based on recurrent selection. Families were derived from open-pollinated seed from a wide range of apple cultivars, as well as crab apples (*Malus* spp.), contributed from repositories from different countries (Notion et al., 1999). Adapted recurrent selection strategies such as those applied in New Zealand can be considered amongst the most promising strategies to keep and increase genetic variability.

At Agroscope FAW Wädenswil we have established a small project to broaden the genetic basis in apple breeding. Diallele crosses were performed including the following cultivars and selections: ‘ZitronenreINETte’, ‘Oberrieder GlanzreINETte’, ‘Discovery’, ‘Delbard Jubilé’, ‘Champagner ReINETte’, ‘Pilot’, ‘Angold’, ‘Reglindis’, A 810-390 (*Pl*<sub>2</sub>), ‘Schweizer Orangenapfel’, HL 75-27 (‘Goldspur’ x ‘Antonovka’), HL 10-04 (‘Goldspur’ x ‘Glockenapfel’) and ‘Nabella’. No major resistance gene against apple scab was included. The purpose of these crosses was to increase the genetic basis with respect to fruit quality and disease resistance. A first seedling is being selected for advanced testing: FAW 13959 (‘ZitronenreINETte’ x ‘Discovery’).

### **Strategies for durable disease resistance**

The challenge of breeding durable disease resistant varieties can be approached in different ways. One promising strategy are molecular techniques that allow to detect the combined presence of resistance genetic factors in a seedling. The availability of molecular markers and genetic linkage maps enables the detection and the analysis of major resistance genes as well as of quantitative trait loci (QTL) contributing to the resistance of a genotype (Liebhard et al., 2003). A promising reduction of the risk of resistance breakdown is the combination of several functionally different

resistances in a cultivar. Examples for molecular selection towards genotypes with such pyramided genetic resistance against scab (*Venturia inaequalis*) and powdery mildew (*Podosphaera leucotricha*) are promising. Molecular markers are available which allow detecting the *Vf*, *Vr* and *Vbj* scab resistance (Tartarini et al., 1999; Hemmat et al., 2002; Gygax et al., 2004) and the *Pl<sub>1</sub>*, *Pl<sub>2</sub>*, *Pl<sub>d</sub>* and *Pl<sub>w</sub>* mildew resistance (Markussen et al., 1995; Seglias and Gessler, 1997; James and Evans, 2004). At Agroscope FAW Wädenswil, we achieved progress in marker-assisted selection by establishing a mass-selection (Frey et al., 2004) and by further reducing cost. We are analysing up to eight markers in a single multiplex reaction. Systems have been established that allow to switch from a SCAR-based screening to a microsatellite based screening on a automated fragment analyzer.

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 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). It is known that the *Vf* scab resistance widely used in apple breeding programmes can be overcome by specific races or strains of the fungus. During this research very diverse and complex resistance behaviours have been found: the cultivars which showed the widest range of resistance were mostly local cultivars and some newly selected hybrids combining major genes and partial resistance.

To achieve a comparable level of resistance as with major genes, several quantitative resistance loci would have to be combined. Liebhard et al., 2003 performed a QTL analysis based on a genetic linkage map that was constructed by using a segregating population of the cross between the apple cultivars Fiesta and Discovery. The progeny was observed for three years at three different sites in Switzerland and field resistance against apple scab was assessed. The QTL analysis revealed 8 genomic regions where six conferred resistance against leaf scab and two conferring resistance against fruit scab. However, the effectiveness of these detected QTL's has to be confirmed at higher disease levels and in other genetic backgrounds.

There is also scope for breeding fireblight resistant apple and pear cultivars by exploiting genetic variation in germplasm and by developing QTL markers. Fire blight, caused by the bacterium *Erwinia amylovora*, is the most serious bacterial disease of pomaceous fruit trees. 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.

## Sources for increased genetic diversity

Some years ago, efforts have been undertaken to enlarge the genetic basis in apple by collecting material in the centers of origin. An expedition to Kazakhstan and Kyrgyzstan by Forsline et al. (1995) was successful. *Malus* collections included 65 accessions (18,000 seeds) representing 3 species endemic to that area. Collection of cuttings of elite materials was kept to a minimum due to restrictions in the USA quarantine facilities. Some unique germplasm from areas that have never been explored was collected. Seven ecosystems were explored in 12 expeditions. In the meantime, this genetic material is being selected for a wide range of fruit and tree characters. The Fruit Genebank at Dresden-Pillnitz (GER) collected about 7000 seeds from 55 accessions as well as scions from 28 accession of *Malus hupehensis*, *Malus kansuensis*, *Malus prattii*, *Malus sieboldii*, *Malus transitoria* and *Malus toringoides* in 2001 during an expedition to 6 sites of the Chinese provinces Sichuan and Chongqing (Geibel and Hohlfeld, 2003).

A national inventory on top and small 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 Agroscope FAW Wädenswil (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.

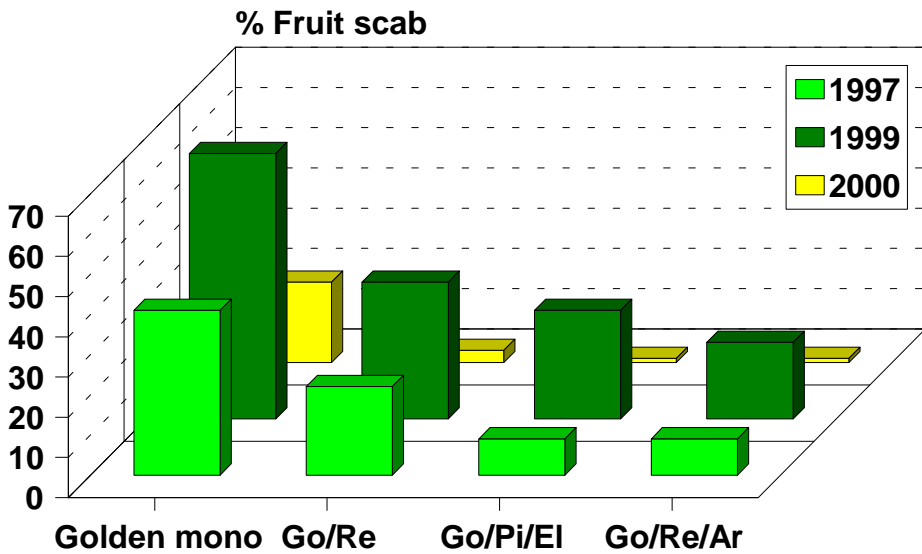
Inventorying, collecting, characterizing, evaluating and utilizing the fruit genetic resources e.g. in a breeding programme are of great public relevance. It allows keeping and utilizing a heritage for future generations, to broaden the genetic basis in breeding programmes and to meet consumers evolving demands of healthy and new and innovative products. The inventory has proven the rich genetic fruit diversity in Switzerland. We have used classical pomological knowledge to verify and determine varietal trueness-to-type. However, many samples remained undetermined, probably being unnamed chance seedlings or unknown varieties. It might be worth considering modern molecular techniques to determine the correct varieties and to highlight synonyms and homonyms (King et al., 1998).

## Nutritional value and health issues

Regular consumption of fruit containing naturally occurring antioxidants has been recommended to interfere in diseases such as cancer and cardiovascular diseases. Stushnoff et al. (2003) have examined juice and fruit tissue from 321 *Malus* species, selections and cultivars from the USDA Plant genetic Resources Unit at Geneva N.Y. A huge diversity in total phenolics and antioxidant capacity was detected. The aim is to supply breeders with data

on antioxidant composition as a guide to parental selection. Information is being gathered on phenols and flavonols to benefit postharvest physiology studies. Data on the compositional diversity of apple germplasm is retained to aid medical epidemiological and dietary intervention studies, as well as identify bioextracts for supplemental application. Fruit will be collected from the designated core collection at Geneva, NY, from wild apples recently collected in Central Asia, and from apple trees in the University of Minnesota germplasm collection.

### Cultivation strategies



**Figure 1.** Effect of apple cultivar mixtures on scab incidence on fruit at harvest of Golden Delicious (Go/Re = ‘Golden’, ‘Rewena’, Go/Pi/EI = ‘Golden’, ‘Pinova’, ‘Elstar’, Go/Re/Ar = ‘Golden’, ‘Rewena’, ‘Ariwa’)

Modern agriculture has led to monocultural production. Monocultures are crops of a single species and a single variety (Phillips and Wolfe, 2004). Durability of resistance is greatly influenced by its management and one of the most convincing systems is based on the concept of mixtures (Blaise and Gessler, 1994). The aim of a research conducted at Agroscope FAW Wädenswil in collaboration with ETH Zürich was to find out if apple cultivar mixtures can reduce the severity of the most harmful apple disease scab. Moreover, effects on powdery mildew, pests and on economy were investigated (Kellerhals et al., 2003). Virulence differences among pathotypes of the scab fungus *V. inaequalis* offer possibilities for the use of host diversity

as a component of disease management. Based on computer simulations, Blaise and Gessler (1994) suggested that apple cultivar mixtures would lead to a reduction of scab epidemics. This hypothesis was tested in an orchard. Tree-wise mixture of three different scab susceptible cultivars ('Elstar', 'Pinova', 'Golden Delicious') significantly reduced scab epidemics on 'Golden Delicious' compared to the pure stand (Fig. 1). Economically the cultivar mixture causes higher labour input which can be partly compensated by reduced cost for plant protection. Due to economical reasons, cultivar mixtures including cultivars derived from genetic resources, can therefore not be generally recommended as a method for improved disease management. However, in particular market situations, if cash flow perspectives (price x quantity) with regard to production costs are favourable or under specific regulations such as organic production, cultivar mixtures may well be a possibility for more sustainable growing systems.

### **Components of sustainable systems**

Sustainability is addressing economical, ecological and social aspects. For agriculture and horticulture, sustainability has been conceptualised more in detail for being able to better address critical issues (e.g. Pretty, 1999; Zachariasse, 2004). The availability of disease and pest resistant cultivars has been identified as a critical bottleneck for making sustainable horticultural systems in the 21<sup>st</sup> century possible (Bertschinger et al., 2004a). The directed use of genetic resources in genetic apple improvement programmes could address many components of sustainable apple growing systems. Durably disease and pest resistant cultivars would be of particular use to organic growing-systems in which pesticide application intensity under temperate climates is even more intense (Bertschinger et al., 2004b) as compared to integrated and conventional systems for the lack of curative pesticides.

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## WYKORZYSTANIE ZASOBÓW GENOWYCH W HODOWLI JABŁONI ORAZ W ZRÓWNOWAŻONEJ PRODUKCJI OWOCÓW

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### S T R E S Z C Z E N I E

W ostatnich latach znacznie wzrosło zaniepokojenie społeczne zmniejszaniem się genetycznej zmienności gatunków roślin uprawnych w świecie. Taka sytuacja sprzyja wspieraniu gromadzenia zasobów genetycznych. Powszechnie wiadomo, że utrzymywanie zmienności genetycznej gatunków roślin sadowniczych umożliwia odpowiedni i nie do zastąpienia zasób genów do poprawy jakości owoców, odporności na choroby i szkodniki przydatnych do hodowli nowych odmian. W pracy analizowane są osiągnięcia i potencjał zasobów genetycznych w nowoczesnej hodowli dla zrównoważonej produkcji jabłek.

**Słowa kluczowe:** hodowla jabłoni, odporność na choroby, zasoby genowe, sadownictwo zrównoważone