

## C Breeding

### C1 Limits and perspectives in plum cultivar breeding using conventional methods

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The breeding work on European plum cultivars (*Prunus domestica* and *Prunus insititia*) is an objective necessity for many countries from Europe, Asia and North America. In this period of beginning of the Millennium, the conventional breeding methods for plum cultivars are still largely used in the majority of programs. The conventional methods have major limitations, which can diminish the perspectives of plum cultivar breeding with consequences on the future of this fruit crop.

The plum breeding programs carried out in Romania during the last 40 years emphasized some aspects regarding the use of conventional methods:

- these methods are still the basis of creating the genetic variability and emphasizing of valuable genotypes;
- continuous modernizing of these methods can create new opportunities and chances of improving the variability;
- require a large volume of work, extended spaces, long selection period, high costs, etc.;
- the modern methods can be complementary to any of the conventional ones (selection, hybridization, ploidy, mutagenesis, etc.).

The conventional breeding methods for plum cultivars are oriented on improving the fruit quality, improving the resistance to diseases, enlarging the adaptability to specific ecological conditions, improving productivity, etc.

### C2 Heritabilities for seedling traits in a *Prunus domestica* L. breeding population

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Heritabilities of seedling growth traits were assessed in a California prune breeding population. A partial diallel design was created with 17 controlled crosses, using 18 parental trees within the UC Davis *Prunus domestica* elite breeding germplasm. The seedlings were grown in a greenhouse for two months followed by ten months in an outdoor nursery planting. Trunk radius, tree height, total branch and main axis length, and growth index (height\*radius<sup>2</sup>) were measured after two months. Trunk radius, tree height, tree fresh weight, and growth index (height\*radius<sup>2</sup>) were measured after ten months.

These data were analyzed using restricted maximum likelihood (REML) to develop estimates of genetic and environmental variances. These variances were used to calculate narrow sense and broad sense heritabilities. Heritabilities differed substantially among traits and were low to moderate in all cases. The utility of these heritability estimates for designing optimal selection strategies depends on the relationship between seedling growth characteristics and traits of commercial importance, and on the consistency of trait expression at different life stages.

### C3 Germination of plum seeds

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When initiating a breeding programme, a proper protocol for seed germination has to be developed. For temperate fruit breeding one has to take into consideration 1) Characteristics of the seed, 2) The medium in which the seed is to be placed and 3) The stratification. In the first years of the Norwegian plum breeding programme, we experienced low and highly variable germination of the seeds. Therefore, we examined different reported methods in order to construct a more efficient protocol, where all these three basic problems were taken into account. The first decision was if embryo, embryo including seed cover or the whole endocarp should be used for the development of a new plant. Several plum breeding programmes utilize embryo rescue techniques in vitro to obtain seedlings, and successful germination with direct sowing of embryos without stratification has also been reported. However, many breeding programmes still make use of stratification of stones, as this is inexpensive and normally gives a reasonable germination. Our experiments demonstrate that several factors are important concerning germination of plum seeds. One of the most critical factors is the control of humidity. A certain loss of germination capacity was also found after drying of seeds followed by water-saturation of embryos and stratification. An experiment with different stratification media will be reported, along with different periods of high and low temperature preceding stratification. The percentage of germination has ranged from 0 to 80% in the experiments, and clearly demonstrates the importance of a defined protocol. After germination, a critical phase of initial seedling growth commences, and the loss of seedlings could be very high.

### C4 Pollination and pollen germination of some new plum cultivars

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Compatibility, amount of pollen and pollen germinability are key factors to be investigated when new cultivars are introduced. The breeder normally informs whether the cultivar is self-fertile or not, and which standard cultivars that may be used as pollinizer. Selection of the best pollinizer are crucial when dealing with self-incompatible plum cultivars, however, the choice of plum cultivars for commercial production is highly different between countries. Therefore, additional pollination experiments are often required upon introduction of new cultivars to a country. In recent years, five new plum cultivars have been recommended for planting in Norway. The first ones to be planted were Jubileum and Reeves, and more recently Avalon, Excalibur and Reine Claude Souffriau. The

presented results are based on obtained fruit set from hybridization in the Norwegian plum breeding programme at Njøs. Pollination was carried out on emasculated and not isolated flowers. Therefore, the results must be regarded as guidelines, as replicated experiments with an orthogonal amount of flowers has not been carried out.

Avalon (Reeves x o.p.) seems to have inherited very poor pollen germination from its ancestor. Both Reeves and Avalon has a very low amount of pollen, and germination was close to zero and hybridization with these two cultivars as the pollen parent gave no fruit set. Excalibur (Cox's Emperor x o.p.) and Reine Claude Souffriau have rich amount of pollen, and good pollen germination. Jubileum (Giant x Yakima) also has a satisfactory amount of pollen and fair pollen fertility. Fruit set in the five cultivars with different pollizers will be tabulated.

## C5 Recent progress in Norwegian plum breeding

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The plum breeding programme at Njøs is quite young. In 1998 Sharka was discovered in our collection of plum cultivars for the first time in Norway. The following years from 1998 to 2001, we were ordered to cut down all fields with observed Sharka. Part of the most valuable material was rescued by using thermotherapy, and subsequent tip-grafting on virus-free rootstocks. In 2001 we started a new series of hybridizations, at a North Carolina design II, with 8 mother cultivars (Edda, Jubileum, Mallard, Opal, Reeves, Reine Claude d'Oullins, River's Early Prolific, Victoria) and 4 pollen cultivars (Early Laxton, Excalibur, Reine Claude Souffriau, Ruth Gerstetter). The crossings have been made in a plastic tunnel on trees trained in super spindle. Crossings in plastic tunnels are made 3-4 weeks before flowering at plain air, where the crossings are made in mobile "tents" covered with agryl. We have experienced low fruit set with Ruth Gerstetter, which was expected as the pollen had poor germinability. On the other hand, Excalibur and Reine Claude Souffriau resulted in high fruit set. Germination of seeds from young trees of Jubileum, Opal, Reeves and Victoria has been high, in contrast to seeds from old trees of Reine Claude d'Oullins and River's Early Prolific.

So far, one selection is tested by growers, 14 selections are tested in our own experimental orchards, and 2000 seedlings are in the process of seedling evaluation.

## **C6 Ripening, quality and resistance donors of genotypes of *Prunus domestica* and their inheritance pattern in the practical plum breeding**

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In more than 30 years of combining breeding of plums and selection of thousands of hybrid progenies of different genetic combinations also the inheritance pattern of heterozygous genotypes of *Prunus domestica* can concentrate in increased and distributed form.

Consequently about the high amount of progenies of conventional combining breeding and with the knowledge of the basic inheritance pattern breeding aims and their successful realisation can arranged better.

Knowing details of practical inheritance pattern is important while genetic engineering has only less public acceptance and also there exist a duty of declaration of fruits.

Genotypes for the breeding parameters maturity time (early, mid and late season), market quality and flavour, disease resistance especial plum pox resistance, shelf life and storability as well as climatic robustness of blue plums and mirabelles are presented and will be characterise in their breeding safeness.

These long-run observations shall also give advices for a specific use of molecular marker tests.

## **C7 Inheritance of Hypersensitivity of European Plum against Plum Pox Virus (PPV)**

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Concerning the economic damage Sharka is the most important virus disease in stone fruit crops in Europe. It is spreading rapidly all over the world. Quarantine measures were only able to decelerate this process. The causing agent of Sharka disease, the Plum Pox Virus (PPV), threatens the cultivation of plum, apricot, peach and, recently, cherry. Up to now, genotypes immune against the virus were not found within the species *Prunus × domestica* L. in spite of great efforts during the past decades. Therefore, previous breeding programs on *Prunus × domestica* aimed at gaining new varieties showing no or only mild symptoms on leaves and, even more important, on fruits when infected with PPV. Some observations made during the last years in different regions of Europe indicate that this quantitative resistance is no longer sufficient to prevent economic damage in plum production as varieties known as quantitative resistant suffer from Sharka, probably due to increasing stress acting on trees. Against the background of this development breeding of plum and prune varieties hypersensitive against PPV becomes more and more important. Hypersensitive genotypes remain healthy in the orchard and are therefore called absolutely resistant against PPV.

Detailed knowledge on the inheritance of hypersensitivity of plum against PPV is a prerequisite for the successful use of this resistance mechanism in breeding programs. The aim of the investigations was to gather information on the inheritance of this trait.

800 genotypes originating from 18 crossing combinations with at least one parent hypersensitive against PPV were tested for their reaction on PPV-infection. Three plants per genotype were grafted on virus free Myrobalane rootstock with a PPV-infected interstem (double grafting). The behaviour of the plants which were grown in the greenhouse for at least four months was monitored. Hypersensitive genotypes showed necrosis on the leaves and on the bark of newly grown shoots as well as tip necrosis. There was quite a broad range of symptoms which can be used as criteria for hypersensitivity.

The percentage of hypersensitive seedlings varied from 0 to 57% depending on the crossing combination. The inheritance of the type of hypersensitivity originating from the Hohenheim gene pool of hypersensitive genotypes (derived from a crossing 'Ortenauer' × 'Stanley', for example the variety 'Jojo' and the number 'Ort×Stan 34') is significantly higher than the one originating from the plum hybrid 'K4' and its descendants. Unexpectedly, crossings between the hypersensitive genotype 'Jojo' and the tasty, but highly Sharka susceptible varieties 'Fellenberg' and 'Felsina' resulted in a high proportion of hypersensitive seedlings. It was shown that resistance against PPV based on hypersensitivity is encoded by genes of the cell nucleus.

Currently, the breeding of plums hypersensitive against PPV is the most promising way to get varieties ensuring the production of plums in Sharka infected regions.