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FINGERPRINTING PEACH VARIETIES USING MOLECULAR MARKERS

prepared by experts from Italy

FINGERPRINTING PEACH VARIETIES USING MOLECULAR MARKERS

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Most of the current peach varieties are characterized by a high level of inbreeding, being derived from a few common ancestral genotypes.

In particular the varieties grown in the United States are virtually all derived from a few accessions that were introduced into North America from Europe or from a single introduction from China known as 'Chinese Cling'.

'Belle of George' and 'Elberta' were originated from open pollination of 'Chinese Cling', and 'J.H.Hale' was originated from open pollination of 'Elberta', which then provided most of the peach cultivars grown in the United States.

'Chinese Cling', 'Elberta' and 'J.H.Hale', which possess high fruit quality, have been extensively used as parents in the peach breeding programs everywhere in the world.

Due to the male sterility of 'Chinese Cling' and 'J.H.Hale' the heterozygosity of the peach varieties has remained appreciably high. But the work of the breeders has resulted in a high level of inbreeding in peach (Scorza *et al.*, 1985; Warburton and Bliss, 1996; Dirlewanger *et al.*, 1998; Testolin *et al.*, 2000; Quarta *et al.*, 2001). In fact, with the widespread practice among breeders to select new cultivars among open-pollinated seedlings, it becomes very close to selection among selfed cultivars. In Peach it is known that open pollination results in less than 5% outcrossing.

At present, the characterization of cultivars is performed by the evaluation of a large set of phenotypic data that is often difficult to assess and it is also necessary to wait for the complete development of the tree before the evaluation. Phenotypic traits are also influenced by the genotype/environment interaction.

Molecular markers could be used in addition to pomological data for variety characterization.

Previous studies using isozymes as markers demonstrated a very low polymorphism among peach varieties due to their common parental genetic background (Arulsekar *et al.*, 1986; Durham *et al.*, 1987; Reynders and Monet, 1987).

Restriction Fragment Length Polymorphism (RFLP) and Random Amplified Polymorphic DNA (RAPD) as molecular markers, were also tested on peach germplasm.

Results obtained on peach by different Scientific Institution using RFLPs, RAPDs and AFLPs (Amplified Fragment Length Polymorphism) will be compared.

If the ideal molecular marker for fingerprinting is one able to produce the largest number of polymorphic bands for each reaction, AFLP seem to be close to the ideal marker since they produce a large number of polymorphic bands.

However, the ideal marker for fingerprinting must, in general, be easy to handle (AFLP are really complex) and highly reliable and reproducible (RAPD are not so reliable). Our experience using RAPD will be also reported.

The dendrogram, obtained by NTYSIS, shows a similarity of about 70% for the Almond 'Ferragnes' with all the other peach varieties and the group of peach x almond hybrids shows results very close to 'Bonanza' and *P.ferganensis*. The similarity index range of the peach genotypes is from 90% to 100% (undiscriminated genotypes).

From the analysis of the pedigree it was possible to explain some of the undiscriminated genotypes. In fact, most of them were natural mutations of the related genotype: 'Redhaven', 'Compact Redhaven' (mutant of 'Redhaven') and 'Redhaven Bianca' (mutant of 'Redhaven');'Springcrest', 'Maycrest' (mutant of 'Springcrest'), 'Dixitime' (mutant of 'Springcrest'), 'Queencrest' (mutant of 'Maycrest') and 'Springbelle' (unknown); 'Armking' and 'Maybelle' (mutant of 'Armking').

'Orion' and 'Venus' were originated from the same cross of 'Stark Redgold' x 'Flamekist'. Both the parents show 'Elberta and J.H.Hale' as common ancestors and several cycles of self or open pollination; the increased inbreeding level makes their discrimination difficult.

'Elegant Lady' and 'Rome Star' show the same pattern despite the different genetic origin. This result cannot be taken as evidence that the two individuals have exactly the same genotype.

'StarkSaturn' and 'Saturne' were confirmed to be the same variety, named differently.

The reliability of RAPD, when submitted to severe conditions, becomes high, even though the total number of polymorphic bands detected for each primer, in some case, decreased of about 50%.

In recent years, Simple Sequence Repeats (SSR) or microsatellites have been increasingly used as markers for fingerprinting, since their reliability is much higher than RAPD and AFLP and their codominance is much more informative compared to the other dominant markers.

Using 26 microsatellites from AC- and AG- enriched libraries (Testolin *et al.*, 2000) most of the undiscriminated genotypes using RAPD also remained undiscriminated, using microsatellites.

Within the group of 'Maycrest' and 'Springcrest' mutants, few genotypes were discriminated. 'Maycrest', which is a sport mutation of 'Springcrest', was discriminated at one locus where 'Springcrest' shows a single fragment and 'Maycrest' carries two alleles. 'Queencrest', which is reported as a mutation of 'Maycrest', showed differences at two loci.

Finally, 'Maybelle' reported as a mutation of 'Armking' was also discriminated at one locus.

The codominance of SSR made it possible to analyze the paternity of several cultivars when one or both parents were included in the sample. In a few cases the official pedigree could not be confirmed. For instance the analysis of 'Starlite' did not confirm 'Springtime' as a

parent. 'Whitecrest', reported as 'Maycrest' mutation, showed differences at ten different loci. It is possible that this was a mislabeled variety.

Dirlewanger *et al.* (in press) demonstrated that the transportability of microsatellites from peach to other *Prunus* species and also to other species that do not belong to the Rosaceae family was very high.

Microsatellites are very powerful markers and are becoming the markers of choice in many plant breeding programs since they are multi-allelic and codominant markers, PCR-based, easily reproducible, randomly and widely distributed along the genome and amenable to automation.

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