

PRODUCTION OF NEW SEEDLESS CULTIVARS FOR MANDARIN IMPROVEMENT

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Seedlessness is one of the most important characteristics for marketing *Citrus* fruit. In the past 10 years, the main goal of the *Citrus* improvement programs developed by CNR IGV-Palermo has been to select new seedless and high quality mandarin cultivars. The production of triploids ($2n=3x=27$) has proved to be a useful and efficient strategy for obtaining seedless cultivars. In order to obtain triploid varieties of mandarin, a wide interploidy cross program has been conducted choosing an autotetraploid ($2n=4x=36$) clone of Dancy tangerine (*Citrus reticulata* B.) as pollen parent and three different diploid ($2n=2x=18$) varieties of mandarin and tangerine (Fortune mandarin, Wilking mandarin and Monreal clementine) characterized by sweet, juicy, easy peeling but, also, seedy fruit.

Crosses were conducted following emasculation of the female parents; the derived fruits were harvested 105 days after pollination for embryo rescue and culture. Flow cytometric analysis (FCM), using a FACStarPLUS flow cytometer and sorter, was used to analyse the relative nuclear DNA content of the cells of the regenerated plants. A molecular marker analysis, inter-simple sequence repeat polymerase chain reaction (ISSR-PCR), provided a definitive characterization of the hybrids obtained.

DNA ploidy evaluation by FCM revealed the presence of triploid plants in all three different progenies obtained. When compared with the diploid and tetraploid controls, the cell lines demonstrated the fluorescence intensity intermediate between the controls, which indicated that they were triploids. Eight triploids were detected in the progeny obtained from the cross between Monreal clementine and Dancy tangerine, ten in the cross Fortune mandarin x Dancy and five in the cross Wilking mandarin x Dancy.

In order to determine the hybrid origin of these progenies, 6 ISSR primers were selected to give polymorphic patterns between the parents of each cross and they were used to analyse the relative offspring. In any case, the ISSR analysis showed specific markers from both parents to segregate in all components of the progeny. In our study, ISSR analysis demonstrated to be a useful and simple method to recognize the hybrid nature of the offspring.

All triploid plants grew very vigorously and have been grafted onto several rootstocks for selection.