

DEVELOPMENT OF AN OLIVE MAPPING POPULATION: *IN VITRO* CULTURE AND SELECTION OF CROSS-DERIVED EMBRYOS BY SSR MARKERS

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Olive (*Olea europaea* L.) is one of the most important species in the Mediterranean basin. Despite its importance, olive has a lack of genetic knowledge and breeding programmes. Nevertheless, up to present time, there is a serious need for developing competitive production systems.

The aim of this experimental work was to develop in short time (six months) an olive population suitable for the construction of genetic linkage map. Traditionally olive germination and seedlings growing take a long time (1-2 years) with an high lost of material (20-30% germination capacity). For these reasons we presented results about an *in vitro* germination protocol and preliminary F₁ paternity test performed directly on F₁ endosperm to select cross-derived embryos. We realized a controlled cross between Coratina and Oliastro and collected 328 F₁ seeds 100 days after blooming time. Seeds were stored in cool chamber at 4°C for 5 months to get over the physiological dormancy of olive. For embryos *in vitro* culture we collected 260 (75%) complete embryos placed individually in sterile test tubes containing 20 ml of hormone free medium in a growth chamber at 19°C with a 16h photoperiod. Germination started after 10 days with an high percentage (85%). During the germination period the contamination of bacteria and fungi was very low (2-3%). After 2 months we started to transfer seedlings in soil in growing chamber at 23°C. The survive percentage was 95%. After 3 months plantlets (12-14 leaves) were transferred in a conditioned green-house.

Subsequently to the evaluation of Coratina self-compatibility (by field analyses), we verified the percentage of cross derived seeds in the F₁ population in order to exclude self-pollinated derived seedlings. To select cross derived embryos, we analyzed the DNA endosperm, recovered from embryos dissection, by SSR markers. We selected the SSR UDO99-039 (Cipriani *et al.*, 2002) that presents a simple profile with any overlapping of parents bands We recognized 80% of embryos originated by cross-pollination and 20% derived by self-pollination of Coratina.