



***In vitro* propagation of olive (*Olea europaea* L.) cv. Koroneiki**

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Abstract

Single node explants of 'Koroneiki' olive trees were cultured for one month on a modified Driver-Kuniyuki for Walnut medium, lacking growth regulators. The explants were subcultured once a month on a medium supplemented with zeatin riboside, 6-(γ - γ -dimethylallylamino) purine, 6-benzyladenine or thidiazuron. Zeatin riboside proved to be superior to other cytokinins in inducing shoot proliferation. The combination of olive knot extract at 25 or 50 mg l⁻¹ with cytokinins suppressed shoot proliferation. After two months at the proliferation stage, the explants were cultured for one week in the dark in 1 ml liquid Woody Plant Medium supplemented with IBA, α -NAA or IBA+ α -NAA. The explants were then transferred to the same solid medium lacking growth regulators, with a small layer of perlite on the surface. The combination of the two auxins at 1+1 mg l⁻¹ resulted in almost 76% rooting. The combination of olive knot extract at 50 mg l⁻¹ with auxins increased the rooting percentage up to almost 87%. Artificial infection of explants with the bacterium *Pseudomonas savastanoi* pv. *savastanoi* inhibited rhizogenesis, even in the presence of auxins. Rooted explants were successfully acclimatized under a mist system, with the survival rate reaching almost 75%.

Abbreviations: ZR – zeatin riboside, 2iP – 6-(γ - γ -dimethylallylamino) purine, BA – 6-benzyladenine, TDZ – thidiazuron, MSN – mean shoot number, MSL – mean shoot length, MNN – mean node number, MNNL – mean node number per shoot length, MRN – mean root number, MRL – mean root length, IBA – indole-3-butyric acid, α -NAA – α -naphthalene acetic acid

Introduction

Micropropagation is a powerful tool for the quick production of genetically homogenous plants, since it enables rapid propagation and hastens the ability of new cultivars. The olive (*Olea europaea* L.) is one of the oldest and most cultivated plants in the Mediterranean basin. It is commonly propagated by rooted suckers, ovules, or by grafting and cutting methods, which usually give a limited number of regenerated plants and great differences in the rooting potential of varieties (Rama and Pontikis 1990). Until today, conventional methods of genetic improvement, such as cross breeding, clonal selection e.t.c. have failed to give satisfactory results, which is mainly due to the

long juvenile period and the high heterozygosity of the species (Rugini 1995).

In vitro culture may overcome some difficulties experienced with conventional propagation techniques and offers an important way for genetic improvement in olive (Rugini (1984, 1995); Rugini et al. 1995). Nevertheless, biotechnological methods, such as *in vitro* culture, require efficient techniques and protocols of *in vitro* proliferation and rooting, particularly when the explants derive from mature tissues.

Reports in the literature on the *in vitro* culture of olive are scant, perhaps on account of the relatively unsatisfactory response of various explant sources to implantation in conventional culture media. Rugini