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Micropropagation of GF-677 rootstocks (*Prunus amygdalus* x *P. persica*)

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SUMMARY – The aim of this research was to facilitate propagation of this hybrid via tissue culture techniques. The explants used in the research were taken from shoot tips in April and sterilized by mercuric chloride. The most suitable medium for culture was modified Knop macroelements. In this medium the use of BA at 1 mg/l level showed the best result for proliferation; and the highest rooting rate was obtained when explants were exposed to LS medium with 0.3 mg/l NAA and 1.6 mg/l thiamin under a 7 day darkness period. Propagated plants were transferred to Jiffy-7 for accumulation and then transferred to pots.

Key word: Micropropagation, tissue culture, proliferation, rooting, shoot tip.

RESUME – *"Micropropagation des porte-greffes GF-677 (*Prunus amygdalus *x* P. persica*)". Le but de cette recherche était de faciliter la propagation de cet hybride par des techniques de culture tissulaire. Les explants utilisés dans cette étude ont été prélevés sur des pousses apicales en avril et stérilisés par chlorure de mercure. Le milieu le plus adapté pour la culture a été modifié par macroéléments Knop. Dans ce milieu l'utilisation de BA à 1 mg/l a donné les meilleurs résultats pour la prolifération, et le taux le plus élevé d'enracinement a été obtenu lorsque les explants ont été exposés à un milieu LS à 0,3 mg/l de NAA et 1,6 mg/l de thiamine sur une période de 7 jours d'obscurité. Les plantes propagées étaient transférées dans des Jiffy-7 pour accumulation et ensuite repiquées en pots.*

Mots-clés : *Micropropagation, culture de tissus, prolifération, enracinement, pousse apicale.*

Introduction

GF-677 is one of the most suitable rootstocks for almond and peach used in calcareous soils to overcome lime-induced chlorosis (Kester, 1970; Fasolo *et al.*, 1987; Hartmann *et al.*, 1990).

This hybrid produces strong roots and has a good potential for pests and diseases (Fasolo *et al.*, 1987; Rathore *et al.*, 1991). The first report about micropropagation of this rootstock was obtained by Kester (1970) and Tabachnik and Kester (1977). Then Fasolo *et al.* (1987) could root explants in the soil. Jona and Gribaudo (1990) successfully measured the amount of ethylen in the medium and final research was accomplished by Dimassi-Theriou (1995) for rooting of GF-677. He compared different culture media and results on the rooting of these rootstocks depend on the type of medium culture (Dimassi-Theriou, 1995).

Material and methods

Different culture media have been used in this research with different hormone concentrations. Other elements were examined to explore the most suitable growing conditions.

Prior to culture, shoots tips containing apical and lateral buds were collected from the trees, carefully transferred to the laboratory and were used as explants. The time period for collecting shoots tips was autumn to spring, although plant material collected on 1st April showed better results.

The best result was obtained with explants sterilized using $HgCl_2$ (0.1%) in 6 minutes. The culture media containing Murashige and Skoog (MS), _ MS and modified Knop (with differences in macroelements) were used in primary stages of the experiment.

Under growth chamber conditions, light intensity was maintained at 2500-3000 lux with an 8-hour dark period. Room temperature and relative humidity (RH) were 24-25°C and 45% respectively.

Explants were transferred to the proliferation medium after 5 weeks and subcultured for every 20 days. Modified Knop was used in the proliferation stage too.

In this stage, hormone treatments contained six levels of BA (0.0, 0.1, 0.4, 0.7, 1 and 2.0 mg/l) and NAA was 0.0, 0.1, 0.2, 0.3 mg/l. The experiment was arranged using a complete design with 10 replications. Data were examined by analysis of variance and the means of treatments were compared using Duncan's multiple test at the 1% level.

In the rooting stage, different culture media were used and their effects on rooting were examined. Note that room temperature was maintained at 25°C during this experimental stage. Propagated plants via tissue culture were transferred to soil medium using: (i) 40% peat and 60% sand mixture; or (ii) using Jiffy-7. Note that room conditions at this stage were the same as in the rooting stage and the only difference was that temperature was reduced from 25°C at rooting stage to 22°C. The initial relative humidity (90%) was gradually reduced for the adaptation of plants to natural conditions.

Results

The results showed that when MS medium was used for culture it caused elongation of explants leaves separation over the culture media. Leaves of these explants showed abscission symptoms which finally caused death of the explants. Good results were obtained using modified Knop media containing 2% sucrose and 75% agar and 1 mg/l BA for proliferation and 0.1 mg/l for elongation shoots (Fig. 1).

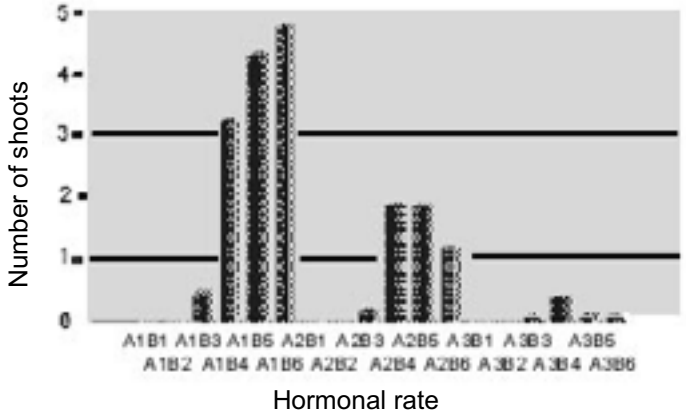

Fig. 1. Interaction effects of NAA and BA on proliferation rate.

The results showed that LS medium containing 0.3 mg/l NAA and 1.6 mg/l thiamin under a 7-day darkness period gave better rooting up to 80% (Fig. 2).

In the transfer-to-soil stage, the using of Jiffy-7 showed better results than the mixture of peat and sand.

Conclusions

The results obtained showed that the addition of more than 1 mg/l BA concentration increased proliferation, but shoots obtained were vitrified.

In the rooting stage, reducing the macroelement concentration to half and increasing phloroglucinol (PG) at levels of 162,320 mg/l did not effect root percentage, but the darkness period

and increase of thiamin were effective for rooting. Also, using NAA more than 0.3 mg/l vitrified explants. It seems that this damage caused by ethylene production in culture medium can overcome by subculture of explants with a period of short.

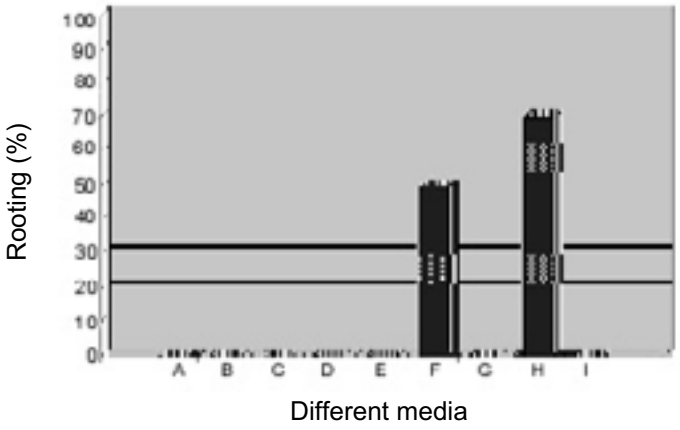

Fig. 2. Comparison of different media.

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