

Cultivar Differences in Shoot Proliferation and Rooting of Apricot (*Prunus armeniaca* L.) in Vitro

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Using seven cultivars of apricot (Heiwa, Mochi-anzu, Yamagata-3, Jinshirou, Shinshu-ohmi, Alfred, and Goldcot), differences in shoot proliferation and rooting ability were investigated. Woody Plant (WP) medium was more effective for the culture establishment of apricot compared with B5 and MS media. High proliferation rates were obtained in 'Alfred' and 'Goldcot', but 'Jinshirou', 'Yamagata-3', and 'Heiwa' gave low rates of shoot proliferation. For shoot length, 'Goldcot' and 'Alfred' gave the best elongation, but poor elongation occurred with 'Yamagata-3', 'Jinshirou', and 'Heiwa'. Rooting ability differed among the four cultivars tested, Heiwa and Mochi-anzu had the highest rooting rate at more than 50%, by comparison a rooting rate of less than 20% occurred in Alfred and Goldcot.

INTRODUCTION

Propagation of apricot (*Prunus armeniaca* L.) cultivars by cuttings has not been successful because they are difficult-to-root species (Snir, 1984). Therefore, they have been propagated by grafting to non-uniform seedling rootstocks resulting in uneven tree growth in the orchard. In vitro propagation is an appropriate method for the mass propagation of clonal rootstocks or own-rooted trees, and results in efficient and uniform orchard management. Recently many kinds of woody plants have been successfully propagated in vitro, but only a few papers have been published on the tissue culture of apricot cultivars (Marino et al., 1993; Snir, 1984). In this study, the differences in micropropagation of apricot cultivars was investigated.

MATERIALS AND METHODS

In late April, about 30-cm-long shoots of seven cultivars (Heiwa, Yamagata-3, Mochi-anzu, Shinshu-ohmi, Jinshirou, Alfred, and Goldcot), collected from mature trees growing under glass, were cut into 3-cm-long segments. These were sterilised by soaking in 70% ethanol for 2 min, immersing in 1% sodium hypochlorite solution for 25 min, and rinsing at least three times in sterile water. Using sterilised scissors they were finally cut into 1-cm-long segments each containing one axillary bud. These explants were used for the following experiments.

Screening Basal Medium to Culture Axillary Buds. Three media [MS, (Murashige and Skoog, 1962), B5 (Gamborg, 1966) and WP, (Lloyd and McCown, 1980)] were used for screening the basal media. Glass tubes (25 mm × 120 mm) with polypropylene caps were used as the culture vessels. Each contained 10 ml of agar-solidified medium. The explants of 'Heiwa' with one axillary bud were planted onto three types of media containing 2 μ M BA, 3% sorbitol, and 0.7% agar at pH 5.8. Unless otherwise stated, the WP medium contained 2 μ M BA, 3% sorbitol, and

0.7% agar. After 30 days of culture, the survival rate of explants and shoot length from axillary buds on each basal medium were recorded. In all experiments, cultures were kept at 26C with a 16-h photoperiod ($40 \mu\text{mol m}^{-2} \text{s}^{-1}$).

Cultivar Differences in Shoot Proliferation and Elongation. Shoots of seven cultivars as described above were proliferated on WP medium and subcultured on the same fresh medium every 3 weeks. Shoots from several subcultures were used for this experiment. In order to evaluate the differences between cultivars in shoot proliferation, shoots of each cultivar were cultured on WP medium. After 30 days of culture, the number of shoots longer than 2 mm and the maximum shoot length were recorded.

In vitro Rooting. The in vitro rooting ability of four apricot cultivars was tested. Shoots 1.5 cm long from subcultures were planted on WP medium containing 0 or $1 \mu\text{M}$ IBA, 3% sorbitol, and 0.7% agar. After 30 days of culture, the rooting rate, and number of roots per rooted shoot in each cultivar were recorded.

RESULTS AND DISCUSSION

Culture Establishment. The survival rate of explants on both WP and B5 media was 100%, while that on MS medium was 84%. The effectiveness of the tested media on shoot elongation from axillary buds was ranked as WP > MS > B5. When shoots from axillary buds were subcultured on the same fresh media, WP and B5 media gave high proliferation rates. However, MS medium gave a low proliferation rate (data not shown). Generally in woody plant species, media supplemented with a low concentration of nitrogen are used for culture establishment (Banno et al., 1989). The nitrogen concentration of WP and B5 media is less than that of full strength MS medium. Therefore, it is considered that nitrogen at high concentrations is not required for culture establishment. Judging from these results, WP medium is the most suitable for the culture establishment of apricots.

Table 1. Cultivar difference in shoot proliferation of apricot.

Cultivar ¹	Number of cultures	Number of shoots	Maximum shoot length (mm)
Mochi-anzu	20	2.6 ± 0.2^2	13.5 ± 0.9
Heiwa	20	1.6 ± 0.2	9.1 ± 0.7
Yamagata-3	20	1.3 ± 0.1	6.8 ± 0.3
Alfred	20	11.0 ± 0.9	16.9 ± 1.3
Goldcot	20	6.5 ± 0.9	17.3 ± 1.6
Jinshirou	20	1.3 ± 0.1	7.2 ± 0.6
Shinshu-ohmi	20	2.5 ± 0.3	11.6 ± 0.6

¹ Each cultivar was cultured on WP medium containing $2 \mu\text{M}$ BA, 3% sorbitol, and 0.7% agar. Data were taken after 30 day-culture.

² Standard error.

Shoot Proliferation and Elongation. The data for shoot proliferation and elongation are shown in Table 1. The shoot proliferation rate and elongation differed among the seven cultivars tested. The shoot proliferation rate ranged from 1.3 to 11.0. 'Alfred' had the highest proliferation, while 'Yamagata-3', 'Heiwa', and 'Jinshirou' gave very low rates ranging from 1.3 to 1.6. Maximum shoot length ranged from 6.8 to 17.3 mm.

Maximum shoot length of 'Goldcot' and 'Alfred' was greater than that of the other five cultivars, while 'Yamagata-3', 'Jinshirou', and 'Heiwa' gave poor elongation.

In Vitro Rooting. The difference in rooting ability is shown in Table 2. 'Mochi-anzu' and 'Heiwa' gave a high rooting rate, over 50%. However, 'Alfred' and 'Goldcot' gave a low rooting rate of less than 20%. No rooting occurred in any cultivar on WP medium without IBA (data not shown). With reference to the number of roots, 'Mochi-anzu' formed three roots, 'Heiwa', 'Alfred', and 'Goldcot' about one root. In many woody species in the Rosaceae (Nemeth, 1986), rooting has depended on genotype. Mature shoots of other woody species are also variable in their response to rooting treatments (Arrilaga et al., 1991). Therefore, similar results might occur in this experiment.

Table 2. Cultivar difference in rooting ability in vitro of apricot.

Cultivar	Number of microcuttings	Rooting (%)	Roots/rooted microcuttings
Mochi-anzu ¹	15	80.0	3.0±0.4 ²
Heiwa	14	57.1	1.5±0.2
Alfred	17	17.6	1.0±0.0
Goldcot	20	25.0	1.2±0.2

¹ Microcuttings in each cultivar were planted on WP medium containing 1 μ M IBA, 3% sorbitol, and 0.7% agar. Data were taken after 30 day-culture.

² Standard error.

From these results, it was found that there were cultivar differences in shoot proliferation and rooting ability. Further experiments are required to clarify optimal conditions for shoot proliferation and rooting.

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Micropropagation of Venus Fly-Trap (*Dionaea muscipula* Ellis)

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Experiments were carried out on a large-scale propagation of Venus fly-trap through leaf explant culture. When whole leaf explants excised from a donor plant grown *in vitro* were cultured on half-strength LS media with different concentrations of BA, adventitious shoots were mainly formed from the petiole of the explants, and few formed from the leaf blade of the explants. The medium supplemented with 2 mg liter⁻¹ BA was the most effective for organogenesis. The shoots grew into plantlets which were transferred to the medium with 0.1 mg liter⁻¹ NAA. The differentiation and subsequent growth of the rhizomes was better in the medium solidified with Gelrite than in that with agar. The adventitious shoots formed in a row on the rhizome in the medium. These shoots were excised from the rhizome and were transferred to the medium for further proliferation. By these procedures, a large number of regenerated plantlets were obtained, and the plants after acclimatization have grown well in pots.

INTRODUCTION

Venus fly-trap (*Dionaea muscipula* Ellis) is an interesting insectivorous plant which belongs to the family Droseraceae. The plant is native to the eastern coast of the United States, and wild species have been reported to be threatened with extinction (Ayensu, 1981). The plant can be used indoors as a potted ornamental plant, and is sometimes used as teaching material for children. The propagation of the plant is usually from seed, however, it is not easy. There are several reports on micropropagation of Venus fly-trap using shoot tips (Hutchinson, 1984), leaves