Adventitious Bud Formation and Plant Regeneration from Leaf Explants of Balloon Flower (*Platycodon grandiflorus*)

M. Kasumi, Y. Takatsu, T. Manabe and M. Hayashi
Plant Biotech Institute Ibaraki, Iwama, Nishi-ibaraki, Ibaraki 319-0292

INTRODUCTION

Until now, adventitious bud formation has not been reported for balloon flower in tissue culture. The objective of the present study was to examine adventitious bud formation and plant regeneration as affected by cultivar, hormone, explant source, and culture conditions with *Platycodon grandflorus*.

MATERIALS AND METHODS

Experiment 1. Effect of plant growth regulators on the efficiency of adventitious bud formation from cotyledon explants was evaluated. Seeds of balloon flower 'Samidaremurasaki' were aseptically sown on Murashige and Skoog (MS) medium. Cotyledons were taken from 15- to 21-day-old seedlings. Cotylendon explants without petioles were cultured on MS medium supplemented with NAA and BAP at various concentrations. Cotyledons were cultured with the abaxial side in contact with the medium. Number of adventitious buds and shoots per explant was reported 60 days after cultivation.

Experiment 2. Effect of organ source on the efficiency of adventitious bud formation was examined. Cotyledons and hypocotyls were taken from 15- to 21-day-old seedlings, and foliage leaves were taken from 30- to 40-day old seedlings. The MS medium was supplemented with 1 mg liter^{-1} NAA and 1 mg liter^{-1} BAP with other culture conditions the same as Experiment 1. Cotyledons and foliage leaves were

Table 1. Effect of plant growth regulators on adventitious bud formation from cotyledons

NAA (mg liter^{-1})	BAP (mg liter^{-1})	No.of explants	No. (%) of adventitous bud		No. of shoots per explant	
0	1	25	19.7 (78.8)	ab	4.0	ab
1	0	25	0.7 (2.8)	d	0.7	d
1	1	25	24.0 (96.0)	a	4.2	a
0	5	25	15.3 (61.2)	b	3.2	b
5	5	25	7.7 (30.8)	c	2.2	c

Means with the same letter do not differ significantly (p=0.05) as indicated by one-way ANOVA followed by Duncan's multiple-range test

cultured with their abaxial side in contact with the medium. The hypocotyls were placed flat on the medium. Culture conditions and experimental procedures were the same as with Experiment 1.

Experiment 3. Cultivar differences in adventitious bud formation were evaluated. Seeds of 'Samidaremurasaki', 'Samidaresiro', 'Shell Pink', and 'Misatomurasaki' were aseptically sown on MS medium. Regeneration medium was the same as used in Experiment 2. Cultures were kept at 20°C under 16-h photoperiod (3000 lux). All experiments were repeated three times.

Figure1. Adventitious bud formation on cotyledon.

Table 2. Effect of kind of organ on adventitious bud formation.

Organs	No.of explants	No. (%) of adventitous bud	No. of shoots per explant
Cotyledon	25	24.3 (97.2) a	4.4 a
Foliage leaf	25	22.3 (89.2) a	3.9 a
Hypocotyl	25	15.7 (62.8) b	3.0 a

Table 3. Varietal difference in adventitious bud formation

Genotypes	No.of explants	No. (%) of adventitous bud	No. of shoots per explant
Samidaremurasaki	25	24.0 (96.0) a	4.2 b
Samidaresiro	25	19.3 (77.2) b	3.2 c
Misatomurasaki	25	25.0 (100.0) a	5.0 a
Shellpink	25	24.0 (96.0) a	4.3 b

RESULTS AND DISCUSSIONS

Experiment 1. Higher adventitious bud formation from cotyledons was observed when MS medium was supplemented with 1 mg liter^{-1} NAA + 1 mg liter^{-1} BAP (96.0% and 78.8%) and 1 mg liter^{-1} BAP, respectively. No significant difference was found between these two treatments. The shoot number showed a similar trend (Table 1).

Experiment 2. Although cotyledons showed the highest adventitious bud formation rate, it was not significantly different from that of leaves. Cotyledon explants produced the largest number of shoots per explant, however, there was no significant difference among organs (Table 2, Fig.1).

Experiment 3. Adventitious bud formation rate was significantly lower for 'Samidaresiro' compared to the other three cultivars with a rate of over 96.0% (Table 3).

Our results demonstrate that balloon flower is able to regenerate plants from cotyledons or true leaves through adventitious bud development on MS medium supplemented with 1 mg liter^{-1} NAA and 1 mg liter^{-1} BAP.