

**High frequency shoots regeneration via direct organogenesis from leaf and petiole of *Solanum aculeatissimum* Jacq.**

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**Objectives**

The objective of the present study was to develop a high-frequency and direct regeneration system for *S. aculeatissimum*, using whole-leaf and petiole explants to compare explants responses to various concentrations of plant growth regulators (PGRs), as well as to determine the optimal media and culture conditions.

**Materials and Methods**

Mature *S. aculeatissimum* seeds, were provided from Bioherb Research Institute, Kangwon National University, Korea.

**Culture condition**

Shoot regeneration potential was examined on different basal media and the effect of carbohydrates sources viz. sucrose, maltose and fructose (10, 20, 30 g/l) were also evaluated for induction of shoots from leaf explants. Different rooting media, MS, 1/2 MS, 1/3 MS, 1/4 MS, MS with different concentration of IAA, IBA, supplemented with 30 g/l sucrose, solidified with 0.8% plant agar were tested.. The origin of the adventitious shoots was studied by means of histological analysis and scanning electron microscopy (SEM).

**Results and Discussion**

The frequency of adventitious shoot regeneration was greatly influenced by the growth regulator concentration and their combination. MS medium supplemented with 0.1 mg/l NAA and 2 mg/l BA demonstrated the best result with higher frequency of shoot regeneration Overall, leaf explants provided better results than those of petiole explants. Among three types of basal salts tested, MS medium supplemented with 0.1 NAA mg/l and 2.0 mg/l BA was effective for higher frequency of regeneration. Histological analysis indicated that shoots originated from the leaf epidermal regions. The morphology of the plantlets was uniform to the seed derived mother plants and they flowered normally within 80 days.

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Table 2. The effect of MS, SH and B5 medium on regeneration of *S. aculeatissimum*.

Medium	Number of shoots/explant	Average shoot length (mm)	% of shoot regeneration
MS	10.33 ± 1.53c	7.33 ± 1.53b	78.67 ± 3.05c
B5	1.67 ± 0.58a	1.17 ± 0.15a	46.00 ± 3.60b
SH	4.00 ± 1.00b	2.47 ± 0.55a	12.33 ± 2.52a

Table 3. Effect of carbohydrate source and concentrations on the number of shoots per explant of *S. aculeatissimum* in regeneration medium.

Carbohydrate source	Concentration (%)	No. of shoot/explant	Shoot length (mm)	% of shoot regeneration
Sucrose	1	1.33 ± 0.58ab	0.70 ± 0.20ab	32.00 ± 2.00g
	2	5.67 ± 1.53d	2.83 ± 0.76de	52.33 ± 2.08h
	3	10.33 ± 2.08e	3.50 ± 0.50d	76.00 ± 3.60i
	4	1.33 ± 0.58ab	1.30 ± 0.20bc	30.67 ± 3.06g
Maltose	1	1.33 ± 0.58ab	1.30 ± 0.20bc	14.33 ± 1.15d
	2	1.67 ± 0.58ab	2.50 ± 0.50de	18.33 ± 0.58e
	3	1.33 ± 0.58ab	1.90 ± 0.96cd	22.33 ± 3.21f
	4	0.00a	0.00a	0.00a
Fructose	1	4.00 ± 1.00c	2.90 ± 0.36de	52.33 ± 2.52h
	2	2.33 ± 1.53b	2.30 ± 1.00de	9.67 ± 2.52c
	3	0.00a	0.00a	0.00a
	4	0.00a	0.00a	0.00a

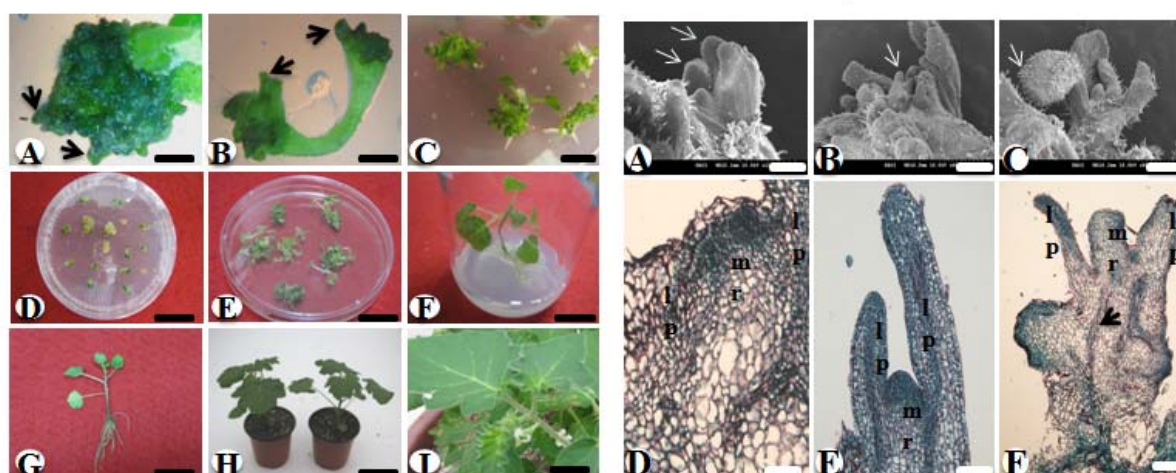


Fig. 1. In vitro multiplication of *S. aculeatissimum*. Fig. 2. Histological and SEM micrograph.