

# **Further Investigation of Australian Native Orange (*Capparis mitchellii*) and Native Lime (*Citrus glauca*) Micropropagation**

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## Abstract

A protocol to micropropagate *Capparis mitchellii* and *Citrus glauca* through plant tissue culture techniques was investigated in this project. An efficient protocol will open a possibility to mass propagate both of these Australian native plants either for industrial or conservation purposes. The project was conducted using surface sterilisation, shoot initiation, shoot multiplication, rooting and acclimatisation experiments.

Microbial contamination level in this project was maintained at a low level by agitating the plant material for 4 hours in 5% (v/v) PPM<sup>TM</sup> solution prior to planting onto media supplemented with mancozeb. By this method, shoot initiation, multiplication, elongation and rooting experiments were made possible.

A range of different strength basal media and growth regulators was utilised to promote shoot and root induction from *C. mitchellii* and *C. glauca* nodal shoot explants. Shoot proliferation was observed in all treatments with the optimum rate obtained on MS media supplemented with 1 mg/L BA and ½MS media with 0.1 mg/L BA for *C. mitchellii* and *C. glauca* respectively. Healthy roots of *C. mitchellii* were promoted by exposing the proliferated shoots to high levels of NAA (100-200 mg/L) for 48 hrs. No roots were observed on *C. glauca* shoots.

Necrotic and secondary abscission zone became a major issue in this project. Prolonged use of antibiotics, imbalance of nutrients and exogenous growth regulators were

considered as the possible factors. Experimenting with nutrient level and growth regulators was necessary to improve shoot and root proliferation.

In conclusion, both *C. mitchellii* and *C. glauca* were propagated by tissue culture methods. This research has provided the first complete protocol to micropropagate *C. mitchellii* and basic information for micropropagating *C. glauca* through nodal shoot culture.

## **Declaration**

I, Syarifah Fadiya Hallaby, certify that this works contain no material which has been accepted for the award of any degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due references has been made in the text.

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## List of Abbreviations

ABA	Abscisic acid;
ANFIL	Australian Native Food Industry Limited
AVH	Australia's Virtual Herbarium
B <sub>5</sub>	Gamborg Plant tissue culture medium
BA	Benzyladenine
BAP	6-benzylaminopurine
CSIRO	Commonwealth Scientific and Industrial Research Organisation
DKW	Driver & Kuniyuki (1984) basal medium for walnut
GA <sub>3</sub>	Gibberellic acid
IAA	Indole 3-acetic acid
IBA	Indole 3-butyric acid;
NAA	$\alpha$ -naphthalene acetic acid;
MS	Murashige and Skoog basal medium
PGR	Plant growth regulators
pH	Potential Hydrogen
PPM <sup>TM</sup>	Plant Preservative Mixture <sup>TM</sup>
TDZ	Thidiazuron
WPM	Lloyd & McCown woody plant basal medium
ZR	Zeatin riboside;