

In vitro cloning of *Actinidia deliciosa* through axillary shoot proliferation and organogenesis

Mariana Neves¹, Filomena Morgado¹, Renato Silva¹, Mónica Zuzarte², Jorge Canhoto¹

¹ Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, 3000-456 Coimbra, Portugal

² Institute for Biomedical Imaging and Life Sciences, Faculty of Medicine, University of Coimbra, 3000-548 Coimbra, Portugal

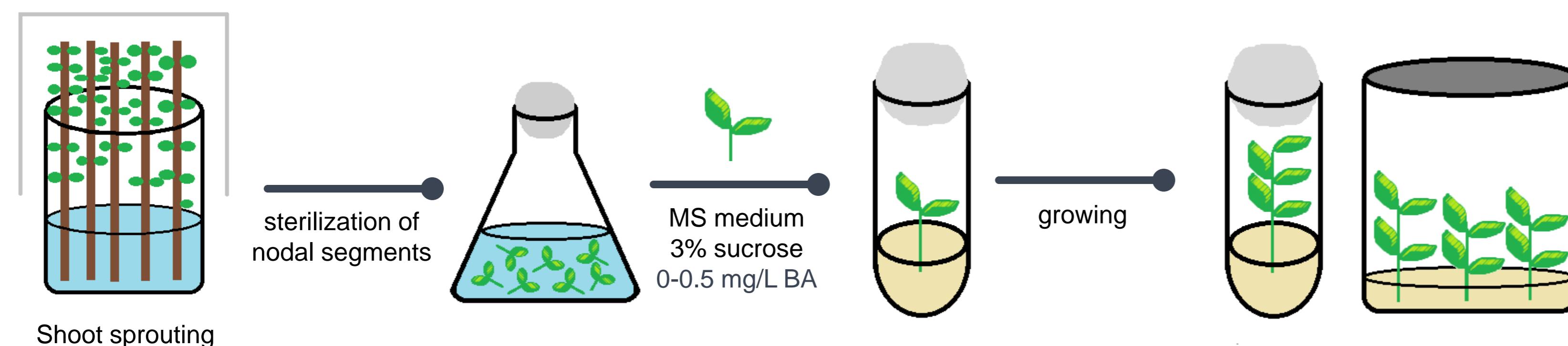
mariananevespt@gmail.com



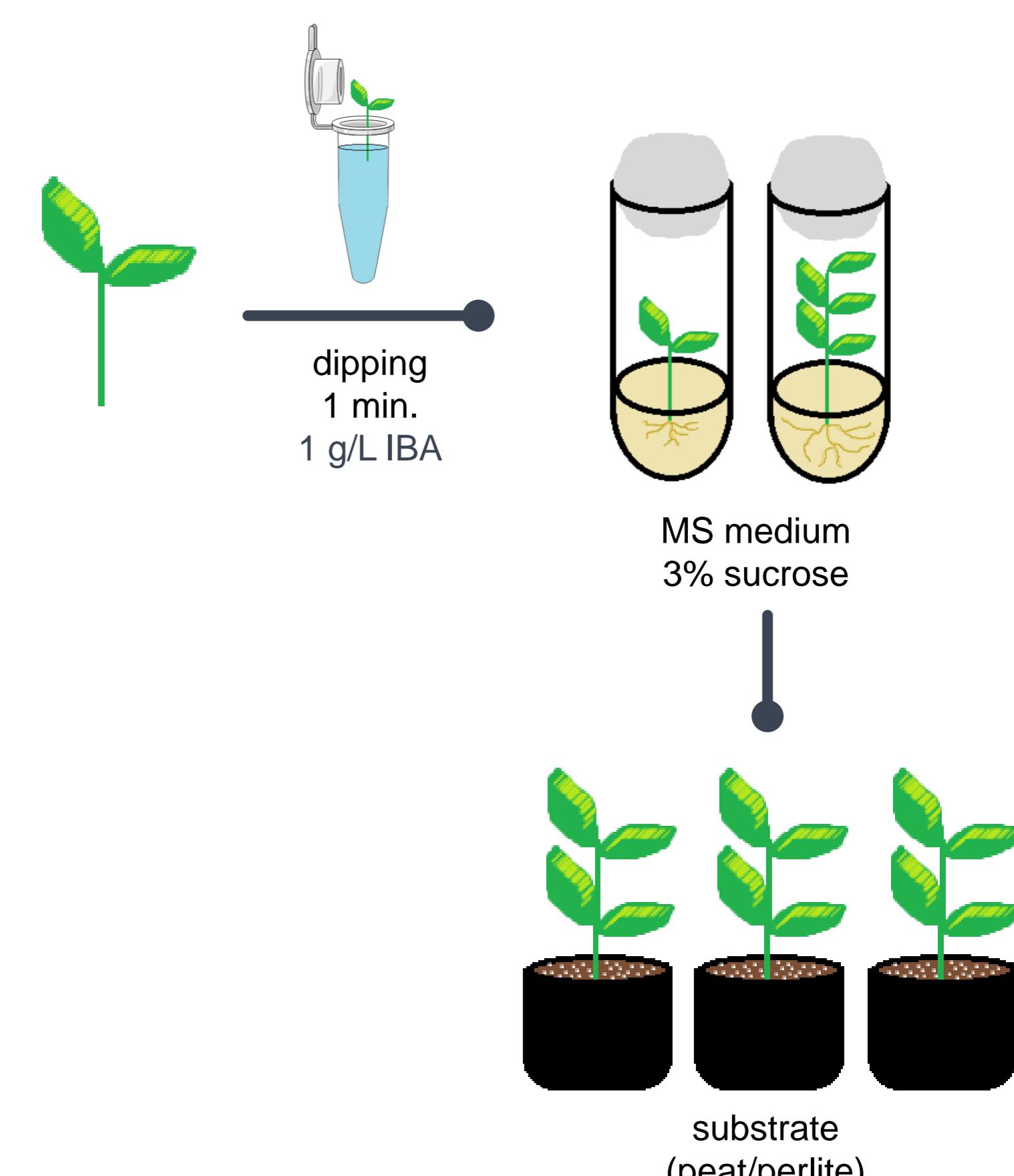
In vitro cloning (micropropagation) can be achieved through different approaches such as axillary shoot proliferation, organogenesis and somatic embryogenesis. These micropropagation methods are high-value biotechnological tools for large-scale cloning propagation and also for the *in vitro* maintenance of clones with interesting characteristics. In this work the first two approaches were applied to clone selected Portuguese plants of kiwi - *Actinidia deliciosa* (A.Chev.) C. F. Liang & A. R. Ferguson - of the female cultivars Hayward and Tsechelidis, and the male cultivar Tomuri. Axillary shoot proliferation was achieved in MS medium containing different concentrations of benzyladenin (BA, 0-0.5mg/L). The sprouts of adult plants' cuttings were also propagated on MS medium with the same range of BA concentration. Nodal segments and apices from established shoots (2-3 cm) were subcultured in the same medium for multiplication. Organogenesis was induced on leaves of proliferating shoots on media containing different combinations of BA (0,2-2.0 mg/L) and NAA (1-naphthaleneaxcetic acid, 0,1 or 0,5 mg/L). Finally rooting was induced *in vitro* or *ex vitro*, using the auxin IBA (indole-3-butyric acid). Using this procedure, several genotypes of the cultivar Hayward and one genotype of the cultivar Tsechelidis were micropropagated.

MATERIAL & METHODS

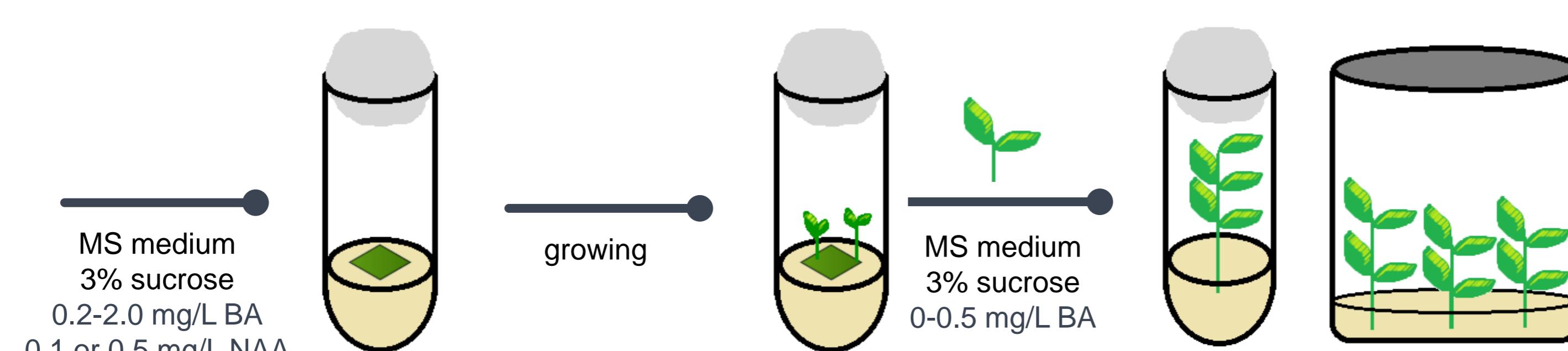
Axillary shoot proliferation



Rooting

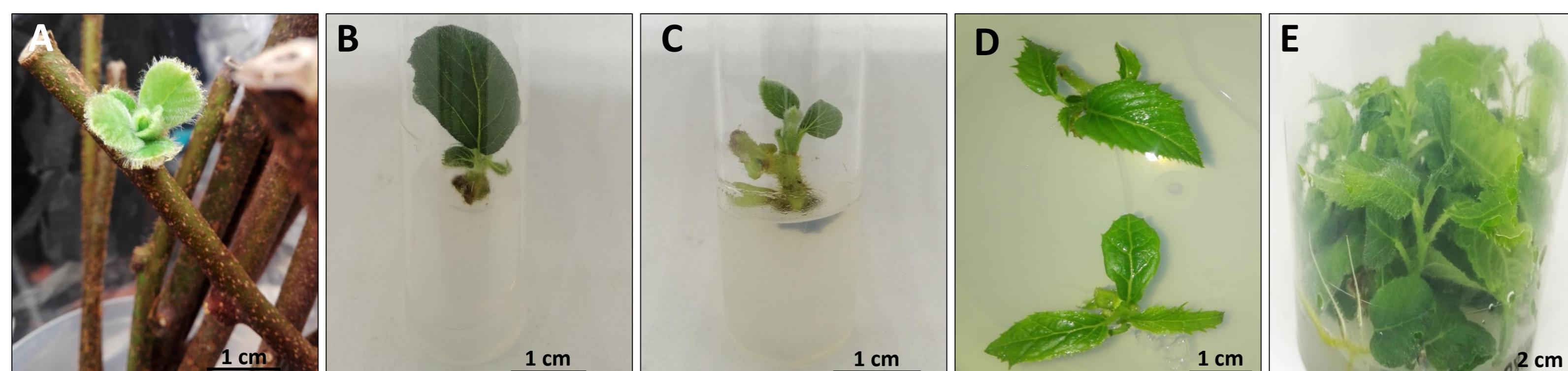


Organogenesis



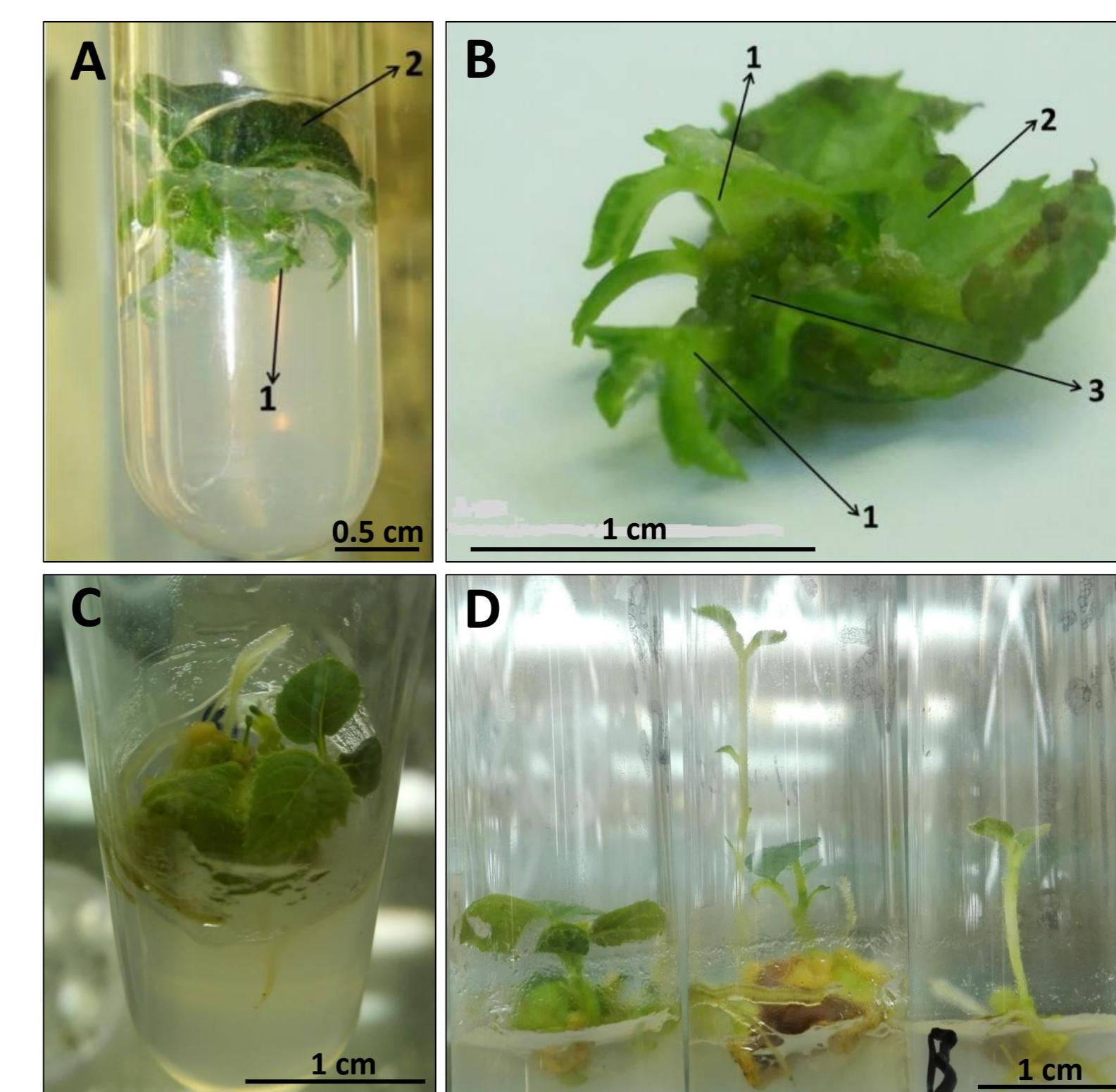
RESULTS

Axillary shoot proliferation



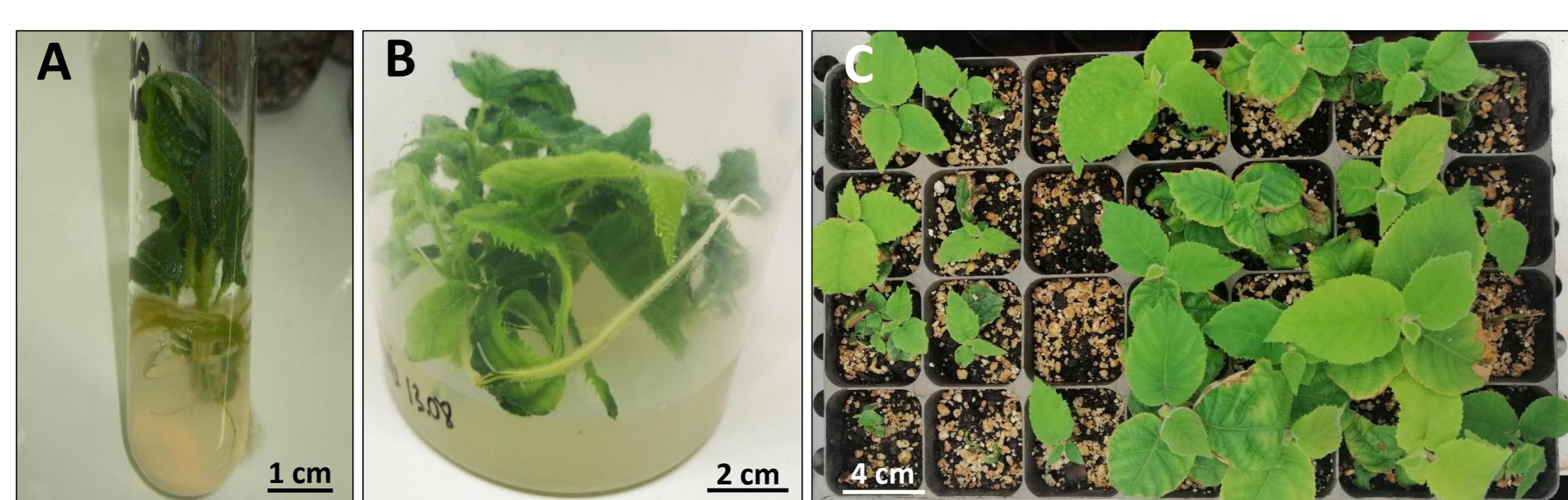
Axillary shoot proliferation. (A) Cuttings (20-30 cm) from field-growing female cultivar Tsechelidis. (B-C) Nodal segments of female cultivar Tsechelidis placed in MS medium containing different concentrations of BA (0-5.0 mg/L). (D-E) Axillary shoot proliferation of the female cultivar (Hayward) and the male cultivar (Tomuri) in MS medium containing different concentrations of BA (0-5.0 mg/L).

Organogenesis



Organogenesis induction. (A-D) Leaf explants with several sprouts. Organogenesis was induced in leaves of proliferating shoots in MS media containing different combinations of BA (0,2-2.0 mg/L) and NAA (0,1 or 0,5 mg/L). (1 - sprout, 2 - leaf expansion, 3 - callus).

In vitro and ex vitro rooting



Rooting induction *in vitro* and *ex vitro* of a female and a male cultivar, Hayward and Tomuri. (A-B) *In vitro* rooting was achieved following a 1 min. dipping in an IBA solution (1 g/L). Shoots were cultured in MS medium for 2-4 week and then transferred to a substrate (peat/perlite). (C) *Ex vitro* rooting was achieved following a similar dipping in the IBA solution and then the shoots were transferred to the same substrate.

CONCLUDING REMARKS

- ✓ Axillary shoot proliferation and organogenesis are effective methods for *in vitro* cloning of different genotypes of *Actinidia deliciosa*.
- ✓ Selected adult plants from the field can be micropropagated through axillary shoot proliferation.
- ✓ Male and female plants exhibit similar *in vitro* responses in both methods.
- ✓ *In vitro* or *ex vitro* rooting seems to be similarly effective methods in this species.
- ✓ Several genotypes of the female cultivars Hayward and Tsechelidis were micropropagated.