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“Filling gaps and removing traps
for sustainable resource management”

In Vitro Conservation and Recovery of *Ullucus Tuberosus* (LOZ.) after Reduced Growth of Microshoots

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Abstract

In vitro conservation by the use of reduced growth is considered to be a reliable biotechnological tool for medium-term conservation of plant germplasm while ensuring its immediate availability. In the present study, we assessed reduced growth condition using five culture media supplements, a cultivation temperature of 4 °C and a 24 h *in vitro* growth in complete darkness, to establish a simple and reliable *in vitro* conservation protocol for ulluco (*Ullucus tuberosus*). Ulluco is an Andean tuberous crop rich in carbohydrates and vitamin C and it represents a staple crop for local people. For this experiment, individual nodal segments of ulluco were precultured for 28 days on half-strength Murashige and Skoog (MS) medium maintained in a culture room under a 16/8 h light/dark regime at 17 °C, and at a photosynthetic photon flux density of 35 $\mu\text{mol m}^{-2} \text{s}^{-1}$ provided by cool-white fluorescent tubes. They were then transferred to half-strength MS medium supplemented with mannitol (10–30 g l⁻¹), sorbitol (10–30 g l⁻¹), sucrose (10–120 g l⁻¹), chlorocholinchlorid (CCC; 300–700 mg l⁻¹) or abscisic acid (ABA; 1–3 mg l⁻¹) and were placed in a cultivation temperature of 4 °C and a 24 h dark conditions for 24 months. Based on survival percentage and number and size of MTs, three superior treatments were selected for further experiment on microtuber germination: mannitol (20 g l⁻¹), sorbitol (30 g l⁻¹) and sucrose (90 g l⁻¹). Three regrowth media were tested: MS, half-strength MS and MS supplemented with 0.5 mg l⁻¹ GA3. After 3-months survival, MT germination and morphological characteristics were evaluated. Results showed that MS cultivation medium supplemented with GA3 and MTs originated from conservation medium supplemented with 90 g l⁻¹ sucrose showed the fastest regrowth and provided overall superior characteristics over plants from other conservation treatments and tested regrowth media. The protocol optimised in this study provides minimal labour and efficient method of ulluco conservation for 24 months.

Keywords: Gibberellic acid, *in vitro* conservation, microtubers, sucrose, ulluco

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