

Storage and high conversion frequency of encapsulated protocorm-like bodies of *Cymbidium devonianum* (orchid)

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Abstract: A method for the storage and high frequency conversion of *Cymbidium devonianum* protocorm-like bodies (PLBs) is reported. To study the effect of nutrient level on storage, PLBs were encapsulated in calcium alginate beads supplemented with 1.0x, 0.5x, 0.25x, or 0.125x Murashige and Skoog (MS) basal medium containing 0.3% (w/v) sucrose, without agar in the encapsulating matrix, and stored at room temperature (25 degrees +/- 2 degrees C) in the dark. Beads containing 0.25x MS were also kept at different temperatures (0 degrees C, 4 degrees C, 8 degrees C, or room temperature) in the dark to ascertain the optimal temperature for storage. One set of controls (i.e., non-encapsulated PLBs) was maintained for each treatment. The survival and subsequent percentage conversion values of PLBs were assessed at 30 d storage intervals after culturing on MS regeneration medium. Compared to the controls, all encapsulated PLBs showed improved storage at room temperature. In all treatments, non-encapsulated PLBs did not survive, but turned brown and died. Encapsulated PLBs in 0.25x MS medium could be stored for 90 d at room temperature without any significant loss in viability. However, a significant decrease in the survival percentage was recorded after longer storage times. Encapsulated PLBs containing 0.25x medium could be stored at 4 degrees C and 8 degrees C for 120 d and 180 d, respectively, without loss of viability. As storage times increased beyond 180 d, the survival percentage of encapsulated PLBs decreased. The conversion of encapsulated PLBs led to the emergence of regenerated plantlets. Initially, small green globular outgrowths from the PLBs were observed on the surface of the beads. These outgrowths multiplied to form clusters of PLBs which then regenerated into plantlets. In both studies, more prolonged storage of encapsulated PLBs increased the time interval for germination and plantlet regeneration on MS regeneration medium. Similarly, decreases in both basal MS strength in the matrix, and in storage temperature, resulted in an increase in the time required for germination and plantlet regeneration. Plantlets that regenerated from stored, encapsulated PLBs were hardened-off, and a high survival percentage (90%) was obtained in a glasshouse.

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