Cryopreservation of protocorm-like bodies (PLBs) of Phalaenopsis bellina (Rchb.f.) christenson by encapsulation-dehydration

ABSTRACT

Protocorm-like bodies (PLBs) of Phalaenopsis bellina were successfully cryopreserved by the encapsulation-dehydration approach. Various stages in obtaining successful cryopreservation using this method were optimized. Encapsulated PLBs precultured in half-strength MS medium supplemented with 0.75 M sucrose for 3 days exhibited the highest viability in terms of 2,3,5-triphenyltetrazoliumchloride (TTC) reduction. The amount of sucrose in the PLBs after incubation in different concentrations of sucrose for different periods of time determined by HPLC. The highest sucrose concentration was 7 mg/g of PLBs for the PLBs treated with 0.75 M sucrose for 3 days as compared to the control which had only 1 mg/g sucrose. After sucrose preculture, the PLBs were subjected to desiccation using one of two methods. Desiccation using silica gel was more efficient in reducing PLBs moisture content. After 6 h of desiccation, PLBs desiccated using laminar air flow had 43.5% moisture content while for those desiccated using silica gel had 32% moisture content. PLBs desiccated to different moisture contents were plunged into LN. After storage in LN the encapsulated PLBs were re-warmed. Two weeks after re-warming PLBs viability was determined by TTC reduction and re-growth assessed. Encapsulated PLBs precultured with 0.75 M sucrose for 3 days followed by desiccated using silica gel for 5 h resulting in a moisture content of 39% lead to the highest post re-warming viability in terms of TTC reduction (46.6% of control PLBs) and 30% re-growth.

Keyword: Moth orchid; Germplasm conservation; TTC; Sodium alginate HPLC