



CRYOPRESERVATION OF COCONUT (*COCOS NUCIFERA* L.) EMBRYOGENIC CALLUS FROM UNFERTILIZED OVARIES BY ENCAPSULATION-DEHYDRATION; A PRELIMINARY STUDY

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Coconut is a perennial oil crop with a large seed which shows recalcitrant behavior. Coconut seed does not have a dormancy period thus limits the application of conventional storage methods. Coconut germplasm are conserved only in field gene banks, making problems with maintenance, labour, cost, adverse weather, pest and diseases. Cryopreservation is the only viable option available for the long-term conservation of coconut germplasm. In this method, plant tissues are stored at an ultra-low temperature, usually that of liquid nitrogen (-196 °C). As a result, cell division and metabolic activities are arrested and thus, plant material can be stored for an unlimited period of time. The objective of the study was to develop a reliable cryopreservation technique for coconut using unfertilized ovary derived embryogenic callus. Embryogenic calli were induced from unfertilized ovaries excised from immature female flowers of variety Dwarf x Tall hybrid (DxT), cultured on modified Y3 medium and cryopreserved by encapsulation-dehydration method. Calli were encapsulated in sodium alginate beads and pretreated with two sucrose concentrations (0.5 M and 0.75 M) for different durations (1, 2 and 3 days). Before application in liquid nitrogen, calli were subjected to dehydration in silica gel for 8 and 16 hours. Water loss from alginate beads after dehydration was observed and survival and recovery of calli were recorded. According to results, water loss (on fresh water basis) is higher in 0.75 M sucrose than in 0.5 M sucrose. The survival of frozen and unfrozen calli in all treatments which were initially pretreated with 0.5 M sucrose was 100%. At 0.75 M sucrose, survival was 100 % except for the treatment of 16 hours dehydration at 3 days interval (90%). Recovery of both frozen and unfrozen calli was affected by treatments and their interactions at both sucrose levels and recovery was very low. Cryopreserved calli pretreated with either 0.5 M or 0.75 M sucrose for one day and dehydrated for 16 h showed 10 % recovery. Overall, the highest recovery frequency was (20%) observed when the calli were pretreated with 0.75 M sucrose for three days and dehydrated for 8 h.

Keywords: *Coconut, Unfertilized ovary, Callus, Cryopreservation, Encapsulation-dehydration*