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Lodgepole Pine: The First Evidence of Seed Based Somatic Embryogenesis and The Expression of Embryogenesis Marker Genes in Shoot Bud Cultures of Adult Trees[1]

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ABSTRACT

Of the various alternatives for cloning elite conifers, somatic embryogenesis (SE) appears to be the best option. In recent years, significant areas of lodgepole pine (*Pinus contorta*) forest have been devastated by the mountain pine beetle (MPB) in Western Canada. In an attempt to establish an SE propagation system for MPB resistant lodgepole pine, several families displaying varying levels of resistance were selected for experimentation involving shoot bud and immature seed explants. In bud cultures, eight embryogenic lines were induced from two of 15 genotypes following various treatments. Genotype had an important influence on embryogenic culture initiation, and this effect was consistent over time. These embryogenic lines were identified by microscopic observations and genetic markers. Despite the abundance of early somatic embryos, the cultures are yet to develop into mature embryos. In contrast, immature zygotic embryos (ZE) cultured from megagametophytes initiated SE at an early dominance stage via nodule-type callus in 1 of 10 genotypes. As part of the study, putative embryogenesis specific genes, WOX2 and HAP3A, were analyzed in cultures of both shoot bud explants and ZE. Based on these analyses, we postulate that PcHAP3A expresses mainly in callus, and may be involved in cell division, while WOX2 was expressed mainly in EM-like tissues. The findings from this study, based on the molecular assessment, suggest that the cell lines derived from bud cultures were truly embryonal mass. Moreover, these experimental observations suggest that PcWOX2 could be used as early genetic marker to discriminate embryogenic cultures from callus.

Keywords : *Stem cell, Somatic embryogenesis, Pine, Mountain pine beetle*

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