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RESEARCH PAPER

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Deterioration of western redcedar (*Thuja plicata* Donn ex D. Don) seeds: protein oxidation and *in vivo* NMR monitoring of storage oils

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Abstract

Deterioration of conifer seeds during prolonged storage has a negative impact on reforestation and gene conservation efforts. Western redcedar (Thuja plicata Donn ex D. Don) is a species of tremendous value to the forest industry. The seeds of this species are particularly prone to viability losses during long-term storage. Reliable tools to assess losses in seed viability during storage and their underlying causes, as well as the development of methods to prevent storage-related deterioration of seeds are needed by the forest industry. In this work, various imaging methods and biochemical analyses were applied to study deterioration of western redcedar seeds. Seedlots that exhibited poor germination performance, i.e. those that had experienced the greatest losses of viability during prolonged storage, exhibited greater abundance of oxidized proteins, detected by protein oxidation assays, and more pronounced changes in their in vivo ¹³C NMR spectra, most likely due to storage oil oxidation. The proportion of oxidized proteins also increased when seeds were subjected to accelerated ageing treatments. Detection of oxidized oils and proteins may constitute a reliable and useful tool for the forest industry.

Key words: Conifer seeds, *in vivo* NMR spectroscopy, MRI, oil peroxidation, protein carbonylation, seed deterioration, seed storage, storage lipids, western redcedar.

Introduction

Successful operation of tree seed nurseries relies on the supply of high quality conifer seeds. Conifer seeds, collected from cultivated or wild stands, are usually stored in tree seed banks for extended periods, and supplied to tree nurseries when requested. In British Columbia (BC), Canada, the Tree Seed Centre (Surrey, BC) is a central facility for seed storage for the entire province. Seeds of several conifer species generally exhibit good long-term storability at -20 °C; nevertheless, even under optimal storage conditions, conifer seeds will deteriorate with time reducing the quality of seedlots, and there are particular species which appear to be especially susceptible to deterioration. Efforts to minimize seed deterioration, are especially important when dealing with valuable or rare seedlots, and seedlots deposited in the seed banks for gene conservation purposes.

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Lipid peroxidation is considered to be a major cause of seed deterioration during prolonged storage (reviewed in McDonald, 1999). The mechanism of lipid peroxidation is relatively well characterized (Hendry, 1993; Frankel, 2005); free radicals attack the unsaturated fatty acids of membrane phospholipids. A decline of seed viability may be related to the peroxidation of phospholipids and consequent membrane damage. Loss of membrane integrity is apparent when excessive electrolyte leakage accompanies seed imbibition (Bewley, 1986). The free radicals generated by membrane damage may subsequently attack other subcellular structures in seeds, including organellar membranes, proteins, and DNA.

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