Notice of the Final Oral Examination
for the Degree of Master of Science

of

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MBiolSci (University of Sheffield, 2012)

“Investigating Potential Physiological Roles of Condensed Tannins in Roots of Populus: Localization and Distribution in Relation to Nutrient ion Uptake”

Department of Biology

Thursday, November 5, 2015
1:00 P.M.
Human and Social Development Building
Room A264

Supervisory Committee:
Dr. Peter Constabel, Department of Biology, University of Victoria (Co-Supervisor)
Dr. Barbara Hawkins, Department of Biology, University of Victoria (Co-Supervisor)
Dr. Patrick von Aderkas, Department of Biology, UVic (Member)
Dr. Doug Maynard, Department of Geography, UVic (Outside Member)

External Examiner:
Dr. Shannon Berch, Soil Ecologist, BC Ministry of Environment

Chair of Oral Examination:
Dr. Robert Dalton, Department of Curriculum & Instruction, UVic

Dr. David Capson, Dean, Faculty of Graduate Studies
Abstract

Condensed tannins or proanthocyanidins (CTs) are polymeric flavonoids and common plant secondary metabolites. They are defined by their protein binding capacity, and they also have anti-oxidant and metal chelating properties. Known functions of CTs include anti-herbivore and anti-microbial defenses. CTs also play a role in soil nutrient cycling and decomposition through protein binding and microbial inhibition. To date, chemical ecology studies, especially on CTs, have focussed almost exclusively on foliar chemistry and above-ground ecological interactions. However, CT concentrations in roots can match and far exceed those found in leaves, particularly in woody plant species. This suggests that CTs are also likely to have important ecological and physiological roles below-ground. A review of the literature suggests three potential roles of root CTs: 1) defense against soil pathogens and root herbivores; 2) facilitating adaptation to toxic soils through heavy metal chelation, and; 3) facilitating or modulating nutrient uptake through the binding of nutrient cations. In this thesis the relationship between root CTs and nutrient uptake will be analysed in P. tremula x alba.

Localization of CTs was determined through both quantitative and histochemical techniques. Quantitation of CTs with the 1-butanol:HCl assay (Porter et al., 1986) clearly showed that CT concentration was higher in the white root zone compared to the brown root zone or cork zone. This is inconsistent with the term ‘condensed tannin zone’ that is commonly used to describe the macroscopic brown zone of the root between the white zone and the cork zone in Pinus (Peterson et al., 1999). The tissue specific and cellular localization of CTs in roots was determined using the CT-specific stain, 4-dimethylaminocinnamaldehyde (4-DMACA), on embedded longitudinal and freehand cross sections. These stained sections showed that CTs were present in cells on the root surface, specifically accumulating in the root cap and epidermal cells. CTs were also sporadically present in the cortical cells of the young, white root zones. CT concentration declined as distance from the root tip increased. The pattern of CT distribution, as indicated by intensity of the staining, corresponded directly to the quantitative assay results.

Growing poplars under low nitrogen availability (0.1mM NH4NO3) stimulated higher root CT concentrations than when they were grown under high nitrogen availability (10mM NH4NO3), suggesting a link between nitrogen and CTs. Since CTs are known to bind cations in vivo. I hypothesised that root CTs may modulate or facilitate nutrient uptake by binding positively charged nutrient ions. The Microelectrode Ion Flux Measurement (MIFETM) technique was used to analyse specific fluxes of NH4+, NO3- and Ca2+ at discreet root locations in live poplar roots, and test for spatial correlation with CT localization. This technique was also used to compare the NH4+, NO3- and Ca2+ fluxes in transgenic high-CT and wild-type (low CT) lines of Populus tremula x alba to test the impact of CTs on nutrient uptake directly. No correlation was found between fluxes of NH4+, NO3- or Ca2+ and CT distribution. These data indicate that root CTs do not influence nutrient uptake in Populus. This study provides important information regarding the pattern of CT distribution as well as fundamental information on nutrient uptake in P. tremula x alba roots. It provides important new knowledge that will be used to stimulate investigations on other potential roles of root CTs.