Genetic Diversity in Mountain Hemlock (*Tsuga mertensiana* (Bong.) Carr.)

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Introduction

In many locations where few other conifers survive, mountain hemlock is an important subalpine species as a protective cover for watersheds, as a wildlife habitat, and as a major component in the long-term maintenance of the subalpine environment. It occurs predominantly in coastal areas between 1000 and 1500 m in southern B.C. and Alaska, at lower elevations in extreme northern ranges, and at higher elevations (1200–2100 m) in southeastern BC, northern Idaho, northwestern Montana, and the Cascade mountains. It is subject to increasing harvesting pressures, and no gene-conservation plan or tree improvement program exists. Little is known about the variation within the species, which is widely spread in latitude and which is disjunct in the interior of the province and adjacent U.S.

With current timber harvesting extending further into the subalpine elevations, it is important to obtain and understand the traits and characteristics of high-montane species, their range, diversity, regeneration potential, and success. Since the Green Plan of the National Forest Genetic Resources Centre (NFGRC) initiative placed emphasis on investigations of minor species, this project was devised to assess major aspects of genetic diversity of mountain hemlock in British Columbia.

Objectives

The objectives of the project are to determine:

1. The mating system. This is the vehicle for moving variation from generation to generation, and which determines how variation is structured within and among populations.
2. Reproductive success and germination ecology. The extent of genetic control over dormancy and germination must be established if *ex-situ* conservation is to be evaluated and if regeneration efficiencies are to be maximized. Simulated (artificial) ageing of seeds will provide data to determine if there is any loss of genetic legacy during storage. Germination ecology will be investigated using a thermogradient system (courtesy of and in collaboration with Dr. C.L. Leadem, B.C. Ministry of Forests) that permits several temperatures and dormancy-breaking treatments to be studied simultaneously and synchronously on a variety of seed sources.
3. Adaptive and quantitative seedling attributes:
   a) Quantitative variations in seedling attributes (height, diameter, shoot/root dry weight) will be analyzed for inter- and intra-population variation in morphology and field performance. A provenance trial has been proposed to study interpopulation differences. A common-garden trial will be planted in 1993 to study intrapopulation variation.
   b) Frost-hardiness patterns/environmental-adaptation patterns will be established with a view to preparing seed transfer guidelines.

4. Genic variation, the variation among and within populations at the gene level, will be determined by means of isozymes. Together these data will provide information on qualitative, physiological, and genetic traits of the species.
5. Morphology of foliage, buds, cones, and seeds collected from designated range-wide locations in British Columbia will be analyzed.
6. Genetic relations to other species:
   a) relationship with other conifer species. Reciprocal controlled pollinations will be conducted to assess hybridization with western hemlock (*Tsuga heterophylla* (Rafn.) Sarg.) (re *Tsuga* X *Jeffreyi*) and Sitka spruce (*Picea sitchensis* (Bong.) Carr.); resulting seeds will be sown and studied for putative hybridity.
   b) a surrogate for other organisms, such as insects and fungi.

Methods

Survey of existing knowledge

To assess the state of knowledge of mountain hemlock, an annotated bibliography, with emphasis on species diversity, reproductive biology, regeneration success, and genetics is being compiled (Table 1).

Generating new information

Generation of new information will be carried out on two levels: among trees within (intra) populations, and among (inter) populations. Sources of materials are:

- two stands represented by individual-tree seed collections (approx. 20 individual trees per stand) from southern Vancouver Island, contributed by Canadian Pacific Forest Products (CPFP), and
- 20 bulked, wild-stand seed collections, made by companies and the B.C. Ministry of Forests (MOF), selected to provide good representation of coastal mountain hemlock, plus one (available) interior representative.

Results

Existing knowledge

Approximately 60 published articles on mountain hemlock have been surveyed; most concern growth rates and tariff tables for estimating tree volumes; stand structure; ecology; fire outbreak, disease susceptibility, and regrowth patterns; biomass; seeding habits; container seedling growth; chemotaxonomy; and nutrient cycling. Scattered information on reproductive morphology and reproductive method, natural regeneration, hybridization, and genetic variation was found. The bibliography will be published in 1993.

New information

Seeds from CPFP's two individual-tree stand collections have been analyzed to determine:

- familial variation in germinative parameters (using standard germination tests). This has been completed; data analysis is under way.
- genotype specificity under simulated (accelerated) ageing. This has been completed; data analysis is under way.
- Seedlings have been grown in containers for a common-garden trial to analyze intrapopulation variation in seedling attributes. Height-growth measurements were made throughout the 1992 growing season; destructive sampling is under way for shoot/root dry weights.

Seeds from the MOF bulk lots are being analysed to determine:

- Reproductive success and germination ecology: Preliminary results show mountain hemlock germination to be highly temperature sensitive, being completed in less than 3 weeks at constant 20°C. However, stratification for 12 weeks produced no germination in 2 months at 15–10°C. Two seed sources were used to develop the appropriate protocol for accelerated-ageing tests; 100% RH at 37.5°C was found to give the clearest results. This treatment will be applied to the remainder of the bulk seed sources (MOF), and to the individual-tree lots (CPFP) to determine if differences in resistance to simulated long-term storage exist, and if these differences are genotype-specific. This is essential information when considering *ex-situ* conservation of genetic diversity.
- Adaptive and quantitative seedling traits. Seeds are being prepared for sowing in a nursery to produce seedlings for (a) studies of interpopulation variation in seedling attributes, and for a provenance trial if resources permit, and (b) for stock on which to test frost hardiness.
- Cone crops were poor in 1992 for mountain hemlock (and most other species) and only one additional seed source was found. Seeds are being extracted.
- Cone, bud, and foliar morphology studies, along with associated organisms (insects, fungi), were deferred to 1993.
- Reciprocal cross-pollinations of western to Sitka spruce were carried out, and cones have been collected and seeds are being extracted. Crosses to mountain hemlock are required.

TABLE 1. Mountain hemlock work plan 1992–1997

Study/activity	1992/93	1993/94	1994/95	1995/96	1996/97
1. Literature review	- Basic listing. - Revisions.	- Revisions completed. - Publication.			
2. Germination ecology and reproductive success					
a. Genetic control	- Data collection completed.	- Analysis in progress.	- Report, submit for publication.	- Publication.	
b. (i) Preliminary test for modelling ageing	- Data collection completed and analyzed.	- Conclusion.			
b. (ii) Accelerated ageing tests	- Test inter pops (MOF)/intra pops (CPFP).	- Continue tests. - Data analysis.	- Data analysis. - Report, submit for publication.	- Publication.	
c. Thermogradient system (MOF)	- Four seedlots tested. - Preliminary data analysis. - Four more lots on test.	- Test four more lots (total 12). - Data analysis.	- Report, submit for publication.	- Publication.	
3. Intrapopulation variation	- Nurser (seedlings grown, two pops, CPFP).	- Analysis of seedling attributes (destructive sampling).	- Report?		
	- As above.	- Establish common garden (1+0).	- Measure, analyze.	- Report, submit for publication.	- Publication.
	- Mating system (CPFP materials).	- Data collection.	- Data analysis.	- Report, submit for publication.	- Publication.
4. Interpopulation variation					
a. Morphology and field performance	- Sow 20 bulked seedlots (MOF).	- Quantitative variation in seedling attributes. - Plantation reconnaissance?	- Complete measurements. Analyze. - Transplant? - Plant to field (provenance trial) or common garden?	- Replant mortality. - Measure. - Data analysis. - Report, submit for publication.	- Publication.
b. Adaptation/frost hardiness	- As above.	- Establish protocols for frost-hard. testing (MOF).	- Complete frost-hard. testing (MOF).	- Analyze. - Report (seed transfer).	- Publication.
c. Genetic variation	- Isozymes (CPFP materials).	- Complete data collection.	- Data analysis.	- Report, submit for publication.	- Publication.
5. Cone, bud, and foliar morphology	- Establish protocols—preliminary work.	- Additional sample (see 4a) collection. - Sample prep./measurement (UBC?).	- Complete measurements (UBC?). - Analyze.	- Complete analysis. - Report, submit for publication.	- Publication.
6. Genetic relation to other	- Crosses made. - Cones collected.	- Extract seeds (92/93 crosses). - More crosses. - Collect cones. - Extract seeds.	- Sow seeds. - Nursery. - Measure seedlings.	- Analyze. - Report, submit for publication.	- Publication.