Preserving Eastern Hemlock Gene Pools through *Ex Situ* Plantings

Belinda D. Esham[1], Scott E. Schlarbaum[1], Charles Burnham[3], Russell A. Cox[4], Leonard J. Cronin[5], Joseph Deschenes[6], Roger Dziengeleski[5], Kristine Johnson[7], David E. Osterberg[5], Christopher Prey[6], James R. Rhea[8], Arnold M. Saxton[2], and Glenn Taylor[7]

[1]Department of Forestry, Wildlife and Fisheries and [2]Animal Science;
University of Tennessee, Knoxville;
[3]Massachusetts Department of Conservation and Recreation;
[4]Tennessee Department of Agriculture, Division of Forestry;
[5]Finch, Pruyn & Company, Inc.;
[6]United States Military Academy, West Point, New York, Natural Resources Branch;
[7]National Park Service, Great Smoky Mountains National Park;
[8]USDA Forest Service, State and Private Forestry, Forest Health Protection

ABSTRACT

Preservation of host species germplasm is a critical component in the overall response to destruction by serious exotic forest pests such as hemlock woolly adelgid. In 1995 and 1996, seed collections of eastern hemlock, *Tsuga canadensis* (L.) Carr., were made in the Great Smoky Mountains National Park (in Tennessee and North Carolina), New York, and Massachusetts to establish *ex situ* plantations of eastern hemlock. The seedlings were grown in containers for three to four years or in containers for two years and transplant beds for two years. In the late winter of 2000, four plantations were established New York (3) and Massachusetts (1). A single plantation was established in the Cumberland Mountains in Tennessee in 2001. Two plantations in upstate New York failed after two continuous years of drought coupled with deer browse. The third New York planting, on the U.S. Military Academy, has good survival and growth, probably related to protection from deer browse. The Massachusetts plantations are surviving, but are infested with hemlock woolly adelgid. Survival and growth are satisfactory in the Tennessee plantation. Challenges in seed collection, nursery production, and planting hemlocks were discussed. The surviving plantations will be eventually managed for seed production to restore locally adapted hemlocks to areas where the species has been decimated by the hemlock woolly adelgid.

KEYWORDS

Eastern hemlock, hemlock woolly adelgid, *ex situ* plantings, germplasm preservation.

INTRODUCTION

The *Tsuga* (Endl.) Carr. genus consists of about 10 species of evergreen, conifer trees native to North America, Japan, China, Taiwan, and the Himalayas. In the United States, eastern hemlock, *Tsuga canadensis* (L.) Carr., has a wide range, occurring naturally from New England to the Georgia mountains and as far west as eastern Minnesota. The species is the most shade-tolerant of all the forest tree species in North America (Godman and Lancaster 1990). For this reason, the species often persists in the under-story or mid-story of the forest for years. Eastern hemlock can take 250-300 years to reach maturity and can live up to 800 years. Approximately 2.3 million acres in eastern North America are dominated by hemlock forests (McWilliams and Schmidt 1999). The trees have a wide variety of habitats, but are most commonly associated with moist forest coves and mountain streams in the southern Appalachian mountains.

Eastern hemlock plays an important ecological role in the eastern forest. It provides a dense and valuable habitat for wildlife such as white-tailed deer (*Odcoileus virginianus* Zimmermann), ruffed grouse (*Bonasa umbellus* L.) and wild turkeys (*Meleagris gallopavo* L.). Several breeding birds are thought to depend on the presence of hemlock for their survival, including blackburnian warbler (*Dendroica fusca* Lath.), black-throated green warbler (*Dendroica virens* Lath.), and blue-headed vireo (*Vireo solitarius* Wilson). Eastern hemlock shades mountain streams, which helps maintain the cool temperatures essential for stream health and the presence of certain aquatic species, such as brook trout. Evans (2004) reported that "the average number of aquatic macroinvertebrate taxa found in hemlock streams was 37% greater than that found in hardwood streams (55 versus 40 taxa). Fifteen macroinvertebrate taxa were strongly associated with hemlock streams, and three taxa were found only in hemlock streams."

In the southern Appalachian mountain region, hemlock glades are critical habitat for many salamander species. This region is considered to be the largest center for salamander diversity in the world. Downed hemlocks provide excellent habitat for salamanders, both in rotting wood and under exfoliating bark. Fallen trees are not only beneficial to salamanders, Ducey and Gove (1999) speculate that coarse woody debris may play an integral role in regeneration of eastern hemlock. Losses of older growth, large trees might not only reduce the seed source, but also reduce the conditions favorable for seed germination and survival. Economically, eastern hemlock was once used as a source of tannin for leather (Godman and Lancaster 1990) and has been important in the pulp and paper industry in modern times. Hemlock is also an important ornamental tree species, often grown by the nursery industry.

Eastern and Carolina (*Tsuga caroliniana* Engelm.) hemlock populations are currently threatened by the exotic insect, hemlock woolly adelgid (*Adelges tsugae* Annand.). Although this adelgid was introduced into the United States approximately 75 years ago from Asia (Annand 1924) it has only become problematic to hemlock populations in eastern forests. The adelgid has caused extensive mortality to eastern hemlock over a large extent of its range. In the United States it has been detected from northeastern Georgia to southeastern Maine and west to eastern Tennessee. It damages trees by inserting piercing, sucking mouthparts into the base of needles and extracting plant fluids. There is also speculation that adelgids insert toxic salivary secretions while feeding. Adelgid activity causes needles to turn brown

and drop. Severe infestations can cause death in as few as four years, but trees can often persist for much longer periods of time. The influence of other insects, fungal pathogens, and environmental factors can exacerbate the impact of the adelgid.

Unlike most other coniferous species utilized for pulp and timber, very little is known about hemlock genetics and diversity (cf. Campbell and Schlarbaum 2002). Aside from some genetic plantations in New England that were established in the 1950s (Olson et al. 1959), eastern hemlock has not been included in tree improvement efforts. Correspondingly, no seed orchards have been established.

Although field studies on eastern hemlock genetics have been limited, there have been ecological and biochemical studies on genetic variability in the species. Kessell (1979) addressed the genetic basis for eastern hemlock's distribution. He found two distinct ecotypes of eastern hemlock, a "high response" and a "low response" type, based on external morphology. The high response hemlock is highly moisture sensitive and demonstrates high growth rates and tolerance to low temperatures. The low response hemlock can also be found on moist sites, but is more common on drier slopes. It has a slower growth rate, low sensitivity to moisture, and high sensitivity to temperature. In the northern portion of the range of the tree, the two forms are distinct with very little intermixing. However, in the Allegheny and Southern Appalachian mountains, the forms introgress to produce a multitude of intermediate types, although these populations still have individuals corresponding to the "high" and "low" response eco-types.

Zabinski (1992) tested disjunct populations of eastern hemlock for isozyme variation, but found very few genetic differences. Based on the morphological evidence of Kessell (1979), she recommended that further research on eastern hemlock include a more comprehensive survey of genetic variation that encompasses a range of morphological and physiological differences.

Schaberg et al. (2003) studied rare alleles in eastern hemlock and found that rare alleles either decreased or increased depending on silvicultural treatment. When trees were selectively cut (small and poor form trees were removed), rare alleles decreased. When trees were diameter-limit cut, rare alleles increased. This indicates that if some rare alleles are lost, the species' ability to adapt to environmental changes could be affected. Alternately, if rare alleles increase then the fitness of the stand is compromised.

The threats posed to native trees by exotic, invasive organisms are well recognized, yet most conservation efforts focus on pest management. In the southern Appalachian region, gene conservation or preservation has been minimal (cf. Schlarbaum, et al. 1999; Campbell and Schlarbaum, 2002). American chestnut [*Castanea dentata* (Marsh.) Borkh.], flowering dogwood (*Cornus florida* L.), American beech (*Fagus grandifolia* Ehrh.), and butternut (*Juglans cinerea* L.) are all examples of tree species that have been severely impacted by an exotic organism without systematic efforts toward genetic conservation.

A complete solution to an exotic pest problem involves conservation or preservation of germplasm (Schlarbaum et al. 1999). Reintroduction of a species after the pest problem is controlled should use locally adapted genotypes. *Ex situ* preservation of germplasm is one

method to ensure the availability of locally adapted seedlings. With forest tree species, *ex situ* plantations are desirable because of delay in reproductive maturation in many tree species. An existing plantation that is managed for seed production will provide seeds in the volumes needed for reintroduction much quicker than seedlings from stored seed.

In 1995, a cooperative project was formed to collect seeds of eastern hemlock from different locations to subsequently establish *ex situ* plantations for eventual conversion to seedling seed orchards. The seeds from the orchards will be used in reintroduction of local genotypes into areas where hemlock populations had been extirpated by hemlock woolly adelgids.

MATERIALS AND METHODS

COOPERATORS

Initial contacts of interested parties were made by the USDA Forest Service, State and Private Forestry, Forest Health Protection, in Asheville, North Carolina. Participants included: the University of Tennessee's Tree Improvement Program (UT-TIP); Finch, Pruyn & Co., Inc. (FP); U.S. Military Academy (USMA) at West Point, New York; Massachusetts Department of Conservation and Recreation (MDCR); and Great Smoky Mountains National Park (GSMNP). The UT-TIP coordinated the seed collection, growing of experimental material, plantation design and analyses, and distribution of plantations. Finch, Pruyn & Co., Inc., provided the funding to initiate the project.

SEED COLLECTION

Seed collections of eastern hemlock were made in proximity of Glens Falls, New York (FP), West Point, New York (USMA), Amherst, Massachusetts (MDCR), various locations in eastern Tennessee, and in the Great Smoky Mountains National Park. Open-pollinated progeny were collected in the 1995 and 1996 fall seasons. The cones were sent to the UT-TIP and air-dried. Seeds were extracted and processed according to the Woody Plant Seed Manual (USDA Forest Service 1974). The seeds were germinated the spring seasons following the year of collection. The seeds were sown in small planting trays and lightly covered with vermiculite. When the resulting seedlings reached the cotyledon stage, they were transplanted into Cone-Tainers™ or Root-Trainers™ each containing approximately 5 cu. in. of commercial soil mix and medium grade pearlite enriched with organic matter.

SEEDLING PRODUCTION

Seedlings from the 1995 collection were grown for two years in containers and then transplanted to the East Tennessee State Nursery near Delano, Tennessee. At the nursery, the seedlings were placed in a transplant bed for one year and then moved to another transplant bed for an additional year. The seedlings from the 1996 collection were grown in containers for three to four years.

PLANTATION ESTABLISHMENT

Four plantations were established with a combination of transplanted seedlings and containerized seedlings in 2000. Two plantations were established in open fields by FP near Glens Fall, New York; one planting was established in a black birch/ eastern hemlock forest by MDCR near Amherst, Massachusetts; and one planting was established under a forest canopy on the reservation surrounding the U.S. Military Academy. A fifth plantation was established in 2001 on a cleared field, using stock grown in Cone-Tainers™, in the Cumberland Mountains of eastern Tennessee on the University of Tennessee's Cumberland Forest Experiment Station (UT-TIP). The fifth plantation was established at a later date, as the site was not ready for planting in 2000.

The FP plantation had tree mats placed around each seedling to prevent competition that were periodically mowed. The USMA plantation was fenced to protect against deer browse and rub. The UT-TIP plantation was mowed after establishment and herbicide sprayed around the trees to control competing vegetation. The MACR plantation received no maintenance.

STATISTICAL DESIGN AND ANALYSES

The experimental design for the plantations was an incomplete block using three tree row plots. All plantations received a mixture of seed sources. The number of families varied at each location.

Only the USMA and UT plantations were measured for survival and height growth. The USMA plantation was measured in 2002, 2003, and 2004, while the UT-TIP plantation was measured only in 2004. The MDCR plantation was observed annually (except for 2003) for attack by hemlock woolly adelgid and identification of potential resistant trees. A mixed model analysis (SAS 2002) was conducted on data from both plantations. For the USMA plantation, the two types of seedlings (transplanted and container) were first nested within family in growth and survival analyses and then combined in a second group of analyses. Analyses to detect differences among seed sources were also conducted. The UT plantation was analyzed to detect family differences in survival and growth.

RESULTS AND DISCUSSION

NURSERY GROWTH

Eastern hemlock seedlings grew relatively slowly in comparison to various pine species, necessitating a longer nursery period before plantation establishment. It was observed that the seedlings in Cone-Tainers™ were generally smaller and less dense than seedlings grown in Root-Trainers™. The seedlings from the 1995 collection were transplanted to nursery beds due to increased size. Similar treatment of the seedlings from the 1996 collection did not occur due to lack of resources. The seedlings did respond favorably to transplanting and were much larger and more robust than the containerized seedlings at planting.

The variety of containers used and uneven treatment (i.e., transplant vs. containerized) of the seedlings was due to the project's primary objective of germplasm preservation rather than genetic improvement. Additionally, the resources supporting this project were very limited, and the seedlings were placed in whatever containers were available at the time.

FIELD STUDIES/FUTURE PLANS

The FP plantations initially had good survival and satisfactory growth. A two-year drought coupled with deer browse, however, reduced the survival to approximately 10 percent in each plantation. Surviving trees are generally under 1 foot in height due to repeated deer browse. Tree shelters will be erected around the surviving trees to salvage what is left of the plantation.

The MDCR plantation was not browsed by deer, but became quickly infested with hemlock woolly adelgid. The majority (85-95%) of the hemlock overstory at this site was infested with the pest. Seedlings, both bare root and in plugs, from various locations were received and out-planted in black birch/eastern hemlock forest in Amherst, Massachusetts. The survival rate of containerized seedlings (90.9%) was different than transplanted seedlings (58%). As of December, 2004, the containerized seedlings had 32 seedlings that were not infested (12.0%), and the transplanted seedlings had only 7 seedlings (13.7%) that were not infested with hemlock woolly adelgid.

It is unknown why the survival rate of the containerized seedlings was higher than that of the transplanted seedlings. It is interesting to note that the percentage of seedlings that have not been infested is approximately the same for both types of seedlings. Data on annually infestation was not available for this paper, but it will be analyzed in the future to see if any family was more susceptible to early infestation. The plantation will continued to be observed for adelgid infestation and a decision to protect the study will be made when it is clear that there are no genotypes resistant to hemlock woolly adelgid.

Analyses of the USMA plantation revealed no difference in survival and growth among the seed sources. Family (seedling type) and family differences were both evident in survival, but not growth. A net loss of growth occurred between 2002 and 2003, probably due to drought.

The USMA plantation will continued to be maintained and eventually converted into a seedlings seed orchard. Hemlock woolly adelgids have not yet attacked the plantation, and protection will be afforded when it occurs.

The UT-TIP plantation showed no family differences in survival or height. Deer browse has been moderate, but portions of the plantation have suffered minor damage from all-terrain vehicles. The plantation will be maintained and protected against hemlock woolly adelgid until approximately age 10. At that time, the plantation will be converted into a seedling seed orchard and all non-GSMNP genotypes will be removed.

CONCLUSIONS

Ex situ germplasm preservation in plantations is an expensive venture, particularly when approached by widely separated cooperators. The slow growth of the eastern hemlock seedlings caused an extended nursery phase that required constant maintenance in the growing season. If creating seed orchards that will provide seed for reintroduction efforts is the primary objective, then grafting local genotypes from different watersheds would be a more efficient approach to seed production: grafts would produce seeds much earlier than seedlings. In addition, a grafted orchard could be established on a grid, making maintenance and seed collection easier than in a thinned genetic plantation.

REFERENCES

Annand, P.N. 1924. A new species of *Adelges* (Hemiptera, Phylloxeridae). *Pan-Pacific Entomologist* 1:79-82.

Campbell, F.T. and S.E. Schlarbaum. 2002. Fading forests II: trading away North America's natural heritage. Healing Stones Foundation. 128p.

Ducey, M.J. and J.H. Gove. 1999. Downed wood as seedbed: measurement and management guidelines, pp. 34-42. *In* Proceedings of sustainable management of hemlock ecosystems in eastern North America, ed. K. Shields. Gen. Tech. Rep. NE-267. Radnor, Pennsylvania: U.S. Department of Agriculture, Forest Service, Northeastern Research Station.

Evans, R.A. 2004. Hemlock woolly adelgid and the disintegration of eastern hemlock ecosystems. *Park Science* 22(2): 53-56.

Godman, R.M. and K. Lancaster. 1990. *Tsuga canadensis* (L.) Carr, pp. 604-612. *In* Silvics of North America, Volume 1: Conifers. Agricultural Handbook 654. U.S. Department of Agriculture, Forest Service, Washington, D.C.

Kessell, S.R. 1979. Adaptation and dimorphism in eastern hemlock, *Tsuga canadensis* (L.) Carr. *The American Naturalist* 113(3):333-350.

McWilliams, W.H.; Schmidt, T.L. 1999. Composition, structure, and sustainability of hemlock ecosystems in Eastern North America, pp. 5-10. *In* Proceedings of sustainable management of hemlock ecosystems in eastern North America, ed. K. Shields. Gen. Tech. Rep. NE- 267. Radnor, Pennsylvania: U.S. Department of Agriculture, Forest Service, Northeastern Research Station.

Olson, J. S., F.W. Stearns, and H. Nienstaedt. 1959. Eastern Hemlock Seeds and Seedlings: Response to Photoperiod and Temperature. Bulletin 620, The Connecticut Agricultural Experiment Station, New Haven. 70 p.

Schaberg, P.G., G.J. Hawley, D.H. DeHayes, and S.E. Niensohn. 2003. Silvicultural management and the manipulation of rare alleles, pp. 67-74. *In* Proceedings of the symposium of the North American forest commission, Forest genetic resources and silviculture working groups, and the International Union of Forest Research Organizations (IUFRO).

SAS Institute, Inc. 2002. SAS/Stat Users Guide, Version 9.1. Cary, North Carolina.

Schlarbaum, S.E. R.L. Anderson, and F.T. Campbell. 1999. Control of pests and pathogens. *In* Ecosystem management for sustainability, Principles and practices illustrated by a regional biosphere reserve cooperative, ed. J.D. Peine. Florida: ERE Press LLC. 500 p.

U. S. Department of Agriculture, Forest Service. 1974. Seeds of woody plants in the United States. U. S. Dep. Agric., Handb. 450. 883 p.

Zabinski, C. 1992. Isozyme variation in eastern hemlock. *Can. J. For. Res.* 22:1838-1842.