Forest Genetics 2025



A joint meeting between the

Western Forest Genetics Association

Southern Forest Tree Improvement Committee

Northern Forest Genetics Association

Proceedings for Forest Genetics 2025: the 38th annual Southern Forest Tree Improvement Committee, and Western and Northern Forest Genetics Association Joint Conference

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The papers and abstracts in these proceedings were submitted by the authors as electronic files. Modifications were made in format to provide for consistency and to allow best possible placement of figures and tables. The authors are responsible for the technical content of their respective papers.

Citation for Forest Genetics 2025:

Hamilton, J.A., and Gottlieb, R. (Eds). 2025. Proceedings of Forest Genetics 2025: the 38th annual Southern Forest Tree Improvement Committee, and Western, and Northern Forest Genetics Association Joint Conference; June 23-27. State College, PA. 115 p.

Citation of papers presented in the Forest Genetics 2025 Proceedings:

Authors' Names. 2025. Title of presentation. In: Hamilton, J.A. and Gottlieb, R. (Eds.) Proceedings of Forest Genetics 2025: the 38th annual Southern Forest Tree Improvement Committee, and Western, and Northern Forest Genetics Association Joint Conference; June 23-27. State College, PA. pp. x-y.

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Acknowledgments and Sponsorships

Message from the Chair



Jill Hamilton, PhD
President, WFGA

Forest Genetics 2025 has been the first-ever joint meeting between the Western Forest Genetics Association (WFGA), Southern Forest Tree Improvement Committee (SFTIC), and the Northern Forest Genetics Associations! As president of the WFGA, it has been my pleasure to lead the organization of this first-ever joint conference under the theme '**Harnessing Genetic Diversity for Resilient Forests'**

With over 170 attendees from across the United States, Canada, Europe, and Asia this meeting drew leaders in the field to discuss the role of genetics in forest health, conservation, climate adaptation, breeding, biotechnology, and much more. The indoor sessions included 80 presentations, including six keynote addresses, with all but one followed by a featured student speaker highlighting a diversity of interdisciplinary research approaches and issues critical to the field of forest genetics. The crew at Penn State coordinated the conference (and added a virtual presentation/attendance option at the last moment) that included pre-conference workshops, an oak pangenome post-conference workshop, a career panel, a student mixer, and four field trips to local forest genetics research sites.

Student support and recognition has always been a key component of each of the individual organizations, and it was an honor to come together and share this opportunity for them to shine. 36 students (2 B.S., 10 M.S., and 24 PhD) gave presentations or posters, showing the future of forest genetics and tree improvement is well represented. Thank you to **Andreas Hamann** for organizing the rubric, assignment, and evaluations of our 16 judges. Congratulations to **Michelle Zavala-Paez (Penn State)** for winning the WFGA Critchfield award for Best Overall Oral Presentation ($1,000) and **Edward Carlsson (Skogforsk, Sweden)** for winning the WFGA Critchfield award for Best Overall Poster Presentation ($500). Congratulations to **Diego Del Orbe Matos (Penn State)** for winning the SFTIC Squillace Award for Best Oral Presentation ($300), **Nora Heaphy (University of Vermont)** for winning the SFTIC Zobel Award for Oral Presentation ($300), and **Mary McCafferty (Penn State)**, **Avalon Miller (Penn State)** and **Zane Smith (UTK)**, who all took home van Buijtenen poster awards ($250, $150, $100, respectively).

There are so many people to thank for making this an incredible conference, but I would particularly like to express our thanks and gratitude to **Tomi Dreibelbis, Corinna Fisher,** and **Kelly Otto** from PSU Conference and Institutes for providing all their time and energy as our local conference service leads. In addition, this conference also would not be possible without the dedicated efforts of our Scientific and Local Organizing Committee representing all three regional groups: **Jonathan Degner, Chen Ding, Andreas Hamann, Sara Lipow, Dana Nelson, Carrie Pike, Fred Raley, Beth Roskilly, Tal Shalev, Zane Smith, Meg Staton, Mary Mason, Nicholas Ukrainetz, Jessica Wright, Denise Alving, Eric Burkhart, Sara Fitzsimmons, Jill Hamilton, Mary McCafferty, Alayna Mead, Amanda Penn, and Michelle Zavala-Paez.**

Acknowledgments and Sponsorships

I would like to thank our workshop organizers (**Meg Staton, Jill Wegrzyn, Fikret Isik, and John Carlson**) for volunteering their time to train next generations of forest geneticists in a range of topics. Thanks as well to **Zane Smith and Michelle Zavala-Paez** for organizing an extremely engaging career panel. Special shout out to **Sara Fern Fitzsimmons** and her amazing field trip crew (**Amanda Penn, Alayna Mead, Mary McCafferty, and Denise Alving)** for leading an inspiring variety of field trips. Thank-you to **Kyra LoPiccolo** for leading the organization of a well-attended student mixer and **Mary McCafferty and Patrick Cumbie** for leading our runners in the 10th annual SFTIC Fun Run! For keeping our website organized and up to date we are incredibly grateful to **Chance Stribling** and thank you to **Rush Studio** for our amazing meeting logo. Finally, I am incredibly indebted to our amazing crew of local undergraduate and graduate student helpers who assisted with all aspects of running the meeting From assessment of location 'vibes' to helping with decorations, technology, registration, and much, much more we are incredibly grateful for **Kingston Gearheart, Mary McCafferty, Raquel Gottlieb, Lucia Anderson, Nathan LaDuke, Kyra LoPiccolo, Michelle Zavala-Paez, Jacob Mazza, Alex Moen, and Diego del Orbe Matos.**

It was truly a great week in Happy Valley!

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1:00pm – 5:00pm	North American Quantitative Genetics	107 Forest Resources
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Program at a Glance

Wednesday, June 25th

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7:30am – 8:30am	Registration Desk Open and Breakfast	Lewis Katz Building
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8:35am – 9:25am	Keynote Speaker: Mats Berlin	118 Katz
9:25am – 9:40am	Student Speaker: Nicholas Boyce	118 Katz
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1:00pm – 5:00pm	Field Trips	
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6:00pm – 9:00pm	Student Mixer	Champs Downtown

Thursday, June 26th

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10:00am – 10:30am	Break	Business Atrium
10:30am – 12:00am	Session 4A: Genomics and Biotechnology	107 Business
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Detailed Program

Monday, June 23rd

Pre-workshops

9:00am	TreeSnap/TreeSnapPlots
10:00am	CartogaPlant/TreeGenes
1:00pm	North American Quantitative Genetics Workshop
2:00pm	Eastern Seed Zone Forum
3:00pm	Hemlock Working Group Meeting *Organizer: Vidya Vuruputoor*
3:00pm	Northern Forest Genetics Association Meeting
4:00pm	Opening Reception

Tuesday, June 24th

Opening and Welcome

8:30am	Opening and Welcome *Jill Hamilton, Matt Keefer, Blair Siegfried*
8:55am	Introduction *Meg Staton*
9:00am	Genomic Tools in Forest Health Conservation, Restoration and Breeding *Keynote Speaker: Richard Buggs*
9:50am	Facilitating Brown Spot Needle Blight Resistance Screening for Loblolly Pine (*Pinus taeda)* with artificial inoculations *Student Speaker: Rhys Eshleman*

Detailed Program

10:05am	Break
10:25am	Introduction *Meg Staton*
10:30am	Genomic Tools in Forest Health Conservation, Restoration and Breeding *Keynote Speaker: Caterina Villari*
11:20am	Genomic Selection across Generations in Norway Spruce, Evaluated with Real and Stimulated Data *Student Speaker: Edward Carlsson*
11:35am	Lunch

Session 1A: Forest Health, Pests, and Pathogens

1:30pm	Lingering Hemlock Search Project: We need your help to restore the mighty hemlock *Olivia Hall*
1:45pm	Advancing Genomic Research and Conservation Efforts for a North American Walnut Threatened by a Non-Native Fungal Pathogen *Cynthia Webster*
2:00pm	Structural Variation and Assembly Artifacts in Two *Fagus grandifolia* Genomes Investigating Candidate Genes for Beech Bark Disease Resistance *Arianna McGuire*
2:15pm	Isobornyl Acetate Overdrive: An 'Omics' Lens into the Eastern Hemlock's Seasonal Defense Strategy against the Hemlock Wooly Adelgid *Vidya Vuruputoor*

2:30pm	Turning Invasive Pests Against Themselves- Protecting Trees with Anti-Pest-Specific Treatments *Dana Nelson (Presented by Thomas Michaels)*
2:45pm	An Assay to Accelerate Screening for Resistance to Dutch Elm Disease in American Elm Trees *Cornelia Pinchot Wilson*

Session 1B: Tree Improvement and Breeding

1:30pm	Collaborative Seed Orchard Program to Supplement White Oak (*Quercus alba)* Tree Improvement Efforts *Laura Dewald*
1:45pm	Effects of Abscisic Acid (ABA) on Maturation and Germination of North American Ash (*Fraxinus* spp.) Somatic Embryos *Emma Land*
2:00pm	Performance of Tolerant Population of European Ash in Clonal and Progeny Trials in Sweden *Beatrice Tolio*
2:15pm	A Cost- Effective Approach to Implement Seed Source Trials in the Pacific Northwest *Stuart Olshevski*
2:30pm	Incorporating Genetic Gains into Loblolly Pine Yield Modeling *Yuhui Weng*
2:45pm	An Integrated Approach to Understanding Adventure Root Induction in Recalcitrant Species *Hayling Liang*

Detailed Program

Session 1C: Population and Quantitative Genetics

1:30pm	Hybridization and Adaptation: The Genetic History of Shortleaf Pine *Austin Thomas*
1:45pm	Unraveling Population Structure and Genetic Diversity of Eastern Cottonwood from the U.S. Forest Service Breeding Program *Carlos Rivera*
2:00pm	Chance or Pattern: Distribution of Major Gene Resistance to the Non-Native Pathogen that Causes White Pine Blister Rust *Kelly Kerr*
2:15pm	Environment-Specific Genomic Prediction of Ash Dieback Resistance in European Ash (*Fraxinus excelsior L.)* *Jaroslaw Burczyk*
2:30pm	Shedding Light on the Genetic Architecture of Adaptive Introgression in a Populus Hybrid Zone *Sammy Muraguri*
2:45pm	Can Allele Frequency Differences Between Populations of Loblolly Pines be Used to Select for Drought Tolerance? *Carol Loopstra*
3:00pm	Break

Session 2A: Climate Change and Adaptation

3:30pm	Pine Species Allocation Across Multiple Countries Under Different Climate Scenarios *Ricardo Cavalheiro*
3:45pm	Environmental Drivers of Planted Seedling Mortality in the Pacific Northwest *Tal Shalev*
4:00pm	Introgression as a Buffer Against Maladaptation in a Keystone Northern Forest Tree *Nora Heaphy*

4:15pm	From Seed to Seedling: Adaptation to Climate in Early Life Stage of *Quercus alba* Populations within an Assisted Migration Framework *Laura Leites*
4:30pm	Interactive Effects of Genetics and Environment on Douglas-Fir Seedlings Responses to Drought and Extreme Temperature Conditions *Emily Von Blon*

Session 2B: Tree Improvement and Breeding

3:30pm	Comprehensive Strategies for Managing Gene Diversity and Growth Potential of Crops by Mean of Selective Cone Harvest in a Seed Orchard of *Pinus koraeinsis* *Ye-Ji Kim*
3:45pm	Pollen Viability Testing Methods to Predict Seed Yields in Loblolly Pine *Austin Heine*
4:00pm	Girdling Topgrafted *Pinus taeda* to Induce Strobili Production *Christopher Heim*
4:15pm	Phenology of Flower Receptivity in a *Pinus taeda* Breeding Orchard can be Predicted with Heat Sums *Mackenzie Bowman*
4:30pm	Rotation-Age Assessment of Realized Genetic Gain in Splash Pine *(Pinus elliotti var. elliotti)* *Andrew Sims*
4:45pm	Woody Diversity and Carbon Stock of Cocoa-Based Agroforestry Systems on the Outskirts of Forest Concessions in Southern Cameroon *Dong Etchike*

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4:00pm	Role of Suppressed Recombination in the Adaptive Radiation of the Hawaiian *Mestrosideros*	*Ashkan Shametov*
4:15pm	QuerChaos: Challenges of Conserving a West Texas Red Oak Syngameon	*Adam Black*
4:30pm	Adaptive Traits of Wide-Ranging Provenances of Longleaf Pine (*Pinus palustris Mill.)* Grown in the Northern Edge of its Natural Range: Implications for Conservation, Restoration and Assisted Gene Flow	*Kurt Johnsen*
5:00pm	STFIC Business Meeting	

Wednesday, June 25th

Opening and Welcome

8:30am	Introduction	*Andreas Hamann*
8:35am	Assisted Migration in Practice	*Keynote Speaker: Mats Berlin*
9:25am	A Climate-Informed Seed Selection Tool for North America: NA-SST	*Student Speaker: Nicholas Boyce*

Detailed Program

9:40am	Break
10:05am	Introduction *Andreas Hamann*
10:10am	Climate Change and False Springs: When Mild Winters Become Dangerous Keynote Speaker: *Jessica Savage*
11:00am	The Role of Climate and Interspecific Gene Flow in Stomatal Evolution Across a *Populus* Hybrid Zone Student Speaker: *Michelle Zavala*- Paez
11:15am	Lunch

Thursday, June 26th

Session 3A: Genomics and Biotechnology

8:30am	Genomics Clues to Patagonia's Fate: Local Adaptation and Future Maladaptation in *Nothofagus pumilio* *Jill Sekely*
8:45am	The Role of Telomeres in Long-Lived Trees: Are they a Biomarker for Environmental Stress? *Diego del Orbe Matos*
9:00am	Gene Expression in Eastern Oak Species is Impacted by Soil Nutrient Profile *Denise Alving*
9:15am	A Genomic Portrait of Regional Introgression in *Quercus macrocarpa* *Rebekah Mohn*
9:30am	A Pan-Genomic Portrait of Regional Introgression in Hybrid Poplars *Baxter Worthing*
9:45am	Genetics-Enabled Resilient Lodgepole Pine Tree Improvement Strategies *Charles Chen*

Detailed Program

Session 3B: Symposium on North American Ash

Organizer: Zane Smith, Meg Staton

8:30am	Progress in EAB-Resistance Breeding Programs	*Jennifer Koch*
8:40am	Field Plantings to Assess EAB Resistance in the USFS Ash Breeding Program	*Mary Mason*
8:50am	Genome Size and Ploidy Variation in *Fraxinus*	*Alan Whittemore*
9:00am	The Evolution of Samara Morphology Differences in *Fraxinus nigra*	*Kyra LoPiccolo*
9:10am	Polyploidy and Hybridization among *Fraxinus* Species in the Great Smoky Mountains National Park	*Zane Smith*
9:20am	Much Assembly Required: An Octoploid Genome of Conservation Concern	*Cris Guzman*
9:30am	Integrating Traditional Breeding and Biotechnology for Resilient Urban Ash Trees	*Nathan Maren*
9:40am	Genomics-Driven Monitoring of *Fraxinus latifolia* (Oregon Ash) to inform conservation and EAB-Resistance Breeding	*Anthony Melton*
9:50am	Multi-Year Analysis of the Chemical Defense Deployed by Green Ash (*Fraxinus pennsylvanica)* Against Emerald Ash Borer *(Agrillus planipennis*) and the Changes in Gene Expression Associated with Successful Defense	*Jeanne Romero-Severson*

Detailed Program

Session 3C: Symposium on Molecular Application in Tree Breeding

Organizer: Fikret Isik

8:30am	Genomic Resources for Loblolly Pine Gene Discovery and Breeding	*Gary Peter*
8:50am	Validation of Genomic Selection Across Three Generations in *Pinus Taeda*	*Nasir Shalizi*
9:10am	Pedigree Assignment Using Likelihood Genomic Relationships in a Breeding Population of *Pinus taeda*	*Carla de Oliveira Castro*
9:30am	Advancing Marker-Assisted Selection for Blight Resistance in Hybrid Chestnut	*Dana Nelson (Presented by Austin Thomas)*
10:00am	Break	

Session 4A: Genomics and Biotechnology

10:30am	Genetic and Environmental Drivers of Intraspecific Variation in Tree Leaf Phenology	*Hanna Makowski*
10:45am	Assisted Migration Needs of Northern Red Oak (*Quercus rubra)*	*Carsten Kuelheim*
11:00am	Providing Biological Context for GWAS Results Using EQTL Regulatory and Co-expression networks in *Populus*	*Mengjun Shu*
11:15am	A Preliminary Assessment of the Future Adaptivity of Eastern Black Walnut (*Juglans nigra L.)* Using Population Genomic Variation	*Vikram Chhatre*
11:30am	Ribosomal Gene Evolution in Chestnut: A Cytogenetic Perspective on Adaptation	*Nurul Islam-Faridi*

Detailed Program

Session 4B: Digital Tools, Networks and Models

10:30am	A Deep Learning Approach to Species Distribution Modelling for North American Tree Species *Zach Zimmerman*
10:45am	Enhancing Forest Research with TreeSnap and TreeSnap Plots: Mobile Platforms for Data Collection and Phenotyping *Florence Caldwell*
11:00am	Tracking Environmental Shifts in Tree Breeding: Unsupervised Machine Learning for Macroenvironment Classification *Aurelio Agular (Presented by Ricardo Cavalheiro)*
11:15am	Genetically Directed Decision Network for Assisted Migration of Eastern U.S. Forest Species Using Web-Based Tools *Christina Kleffer*
11:30am	Future-Proofing Forests Through a Genetically Informed Reforestation/Restoration Network *Kevin Potter*

Session 4C: Symposium on Molecular Application in Tree Breeding

Organizer: Fikret Isik

10:30am	Genetics of Dwarfism in *Pinus taeda* *Pina Guner*
10:50am	Gene Stacking for Fusiform Rust Resistance in *Pinus taeda* *Trevor Walker*
11:10am	Genome Characterization and Population Genetic Analysis of *Cronartium Quercuum f.sp. fusiforme* in a Controlled Inoculation Trial *Makayla Rutski*
11:30am	Major QTL Confer Race-Nonspecific Resistance in the Co-evolved *Cronartium quercuum f.sp.fusiforme- Pinus taeda* Pathosystem *Fikret Isik*

Detailed Program

12:00pm	Lunch
12:30pm	Career Panel
2:00pm	Poster Session
4:30pm	WFGA Society Meeting

Friday, June 27th

Session 3A: Genomics and Biotechnology

8:30am	Introduction *Michelle Zavala- Paez*
8:35am	Genetic Variation Underlies Differential Responses to Climate Change in Trees: Predicting and Taking Advantage of Rangewide Heterogeneity *Keynote Speaker: Alayna Mead*
9:25am	Utilizing Southern Oak Genotypes as a Restoration Strategy in Our Northeastern Forests *Student Speaker: Samantha Fuentes-Gigliotti*
9:40am	Break
10:05am	Introduction *Michelle Zavala- Paez*
10:10am	Addressing Conservation Priorities and Knowledge Gaps with Help from Genetics- A Story of Partnerships in Support of Red Spruce Forests *Keynote Speaker: Stephen Keller*
11:00am	Closing/Poster & Speaker Awards
12:35pm	Lunch
1:00pm	Post-Conference Oak PanGenome Workshop and Lunch

Keynote Speakers

Richard Buggs
Queen Mary University of London

A TALE OF TWO PANDEMICS FROM A HOST GENOME PERSPECTIVE

Richard J. A. Buggs [1,2]

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[2] Senior Research Leader (Plant Health and Adaptation), Royal Botanic Gardens Kew, Kew Road, London, TW9 3DS



In the past century Britain has been invaded by Dutch elm disease and ash dieback, devastating its populations of *Ulmus minor* and *Fraxinus excelsior*. I will describe research on the genomic composition of these tree populations before and after invasion. Limited genomic variation was present in British elms pre-pandemic, with many common clones. Although Dutch elm disease is normally fatal to large trees, natural selection is inefficient because trees do not die until ~12 years old, tree roots survive to sucker, and little sexual reproduction occurs. Decades of crossing programmes involving Asiatic elm species have produced a plethora of resistant hybrids and introgressants. These are widely planted in Britain, resulting in higher genomic diversity than pre-pandemic, almost entirely mediated by humans. However, the iconic form of the large "English elm" has not yet been recovered, and the genomic basis of resistance has not been fully characterized. By contrast, pre-pandemic British ash populations harboured high genomic diversity. Natural selection by ash dieback is efficient because it kills juvenile trees and sexual reproduction of ash is abundant. Low susceptibility to ash dieback is a highly polygenic trait and shifts in allele frequencies at thousands of relevant loci have been demonstrated in a natural woodland using genomic prediction methods. Unlike elm, populations of ash in Britain may recover within a few decades by natural means. If not, or if emerald ash borer starts to devastate British ash, hybrid breeding may be necessary for ash too. Studies of molecular convergence have identified candidate genes for resistance to ash dieback and emerald ash borer in Asiatic ash species, which may facilitate breeding or gene editing. Knowledge of population genomics for other tree species may enhance our response to future tree pest and pathogen invasions, allowing swift assessment of the likely outcome and informing management responses.

FACILITATING BROWN SPOT NEEDLE BLIGHT RESISTANCE SCREENING FOR LOBLOLLY PINE (*PINUS TAEDA*) WITH ARTIFICIAL INOCULATIONS

Rhys A. Eshleman [1], Katie McKeever [2], Zachary Bragg [1], M. Nasir Shalizi [3], Trevor Walker [3], and Caterina Villari [1]

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[3] Cooperative Tree Improvement Program – North Carolina State University, Raleigh, NC, USA 27606

Within the past two decades, pine forests have experienced a dramatic increase in the incidence and severity of needle diseases, which cause premature needle-drop and growth reductions. Frequent reports of brown spot needle blight (BSNB; caused by fungal pathogen *Lecanosticta acicola*) on loblolly pine (*Pinus taeda*) have prompted concern among foresters and landowners in the region, especially because loblolly pine was not considered a susceptible species until recently. Breeding could be a potential option to increase resistance against BSNB disease and mitigate the damage. However, breeding is contingent on the genetic control of disease resistance, and there are currently no published estimates of resistance inheritance toward BSNB in loblolly pine. In this project, we developed and optimized a high-throughput greenhouse screening protocol for *P. taeda* seedlings against BSNB and used it to screen 94 commercial open-pollinated families from a variety of provenances. Greenhouse screenings are a valuable tool to pair with field progeny tests, as they have a significantly faster turnaround time and reduce environmental variation, such as uneven disease pressure. Preliminary results show significant genetic variation among families, with a family mean heritability of 0.71. Provenance effects, on the other hand, were not significant. Further experiments, including paired field trials, applied comprehensive screenings, and characterization of host resistance physiology are needed to elucidate *P. taeda* resistance to BSNB.

Keynote Speakers

Caterina Villari
University of Georgia

NEW TECHNOLOGIES TO THE RESCUE: OPTIMIZING THE SELECTION AND DEPLOYMENT OF GENETICALLY IMPROVED TREES

Caterina Villari [1], Simone Lim-Hing [2], Colton Meinecke [1]

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[2] North Carolina State University, Raleigh, NC, USA 27695



Successful tree resistance programs rely on multiple steps, from the initial identification of putative genetically superior trees to the deployment of improved material in optimal areas. Recent advancements in cost-effective technologies and computational methods have the potential to enhance each stage of this process. In this talk, I will present two case studies demonstrating the use of near-infrared (NIR) spectroscopy, machine learning, and new statistical approaches to advance the selection and deployment of disease-resistant loblolly pine families. The first study focuses on resistance to fusiform rust (caused by *Cronartium quercuum* f. sp. *fusiforme*), which is one of the most significant diseases of loblolly pine. We developed a protocol combining NIR spectroscopy with machine-learning approaches to classify resistance phenotypes based on the chemical signature of a given tree, with the goal of improving the accuracy of genotype ratings. Using a hand-held device, our approach achieved up to 69% accuracy in distinguishing highly resistant or susceptible trees across the southeastern United States within seconds. Additionally, we generated new fusiform rust hazard maps to assist in the identification of high-risk areas where the use of improved families is recommended. We first used a Least Absolute Shrinkage and Selection Operator (LASSO) regression to identify relevant predictor variables and then modeled them using Generalized Additive Models (GAMs). Our models were able to explain up to 93.1% of the deviance for disease incidence and demonstrated a difference in the influence of climate on the predicted distribution of the disease based on tree genotype. The second study, which is ongoing, focuses on brown spot needle blight (an emerging disease on loblolly pine caused by *Lecanosticta acicola*) and aims to refine the NIR spectroscopy protocol for use with younger seedlings in greenhouse conditions, facilitating the selection of resistant families at earlier growth stages.

GENOMIC SELECTION ACROSS GENERATIONS OF CLONALLY PROPAGATED NORWAY SPRUCE – EVALUATED ON REAL AND SIMULATED DATA

Edward A. Carlsson [1], Harry X. Wu [2], Henrik R. Hallingbäck [3], Jon Ahlinder [4], Mari Suontama [4`]

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[3] Uppsala Science Park, Skogforsk, SE-75183 Uppsala, Sweden

[4] Department of Tree Breeding, Skogforsk, Box 3, Tomterna 1, Sävar SE-91821, Sweden

The genetic gain from forest tree breeding is hindered by long rotation times due to a combination of late reproductive maturity and the need for rigorous field testing. Genomic selection (GS) aims to accelerate this process by enabling genome wide, marker-based selection in populations without phenotypic data. However, the accuracy of GS must first be thoroughly tested using data from two generations that has been both phenotyped and genotyped. In this study, we evaluated prediction accuracy across two linked generations of clonally propagated Norway spruce (*Picea abies*): one consisting of 954 plus trees (G0) and 956 progeny trees representing 34 full-sib families (G1). These were tested across a total of 16 field trials across mid- and south Sweden. Both generations were measured for height and genotyped using a 50K SNP chip array. We estimated prediction accuracy both across and within generations, considering between- and within-family accuracy. Various cross-validation scenarios were designed to assess the impact of relatedness between training and validation sets on prediction accuracy. Additionally, we used computer simulations with AlphaSimR to emulate the real data and repeat the same analysis across different historical effective population sizes. Cross-generational GS exhibited considerably lower accuracy than within-generation GS, both in real data (0.15–0.29 vs. 0.49–0.76) and simulations (0.18–0.30 vs. 0.59–0.80). Among cross-generational scenarios, accuracy tended to increase as the training set became more closely related to the validation set, despite even a tenfold reduction in training set size. A combination of direct parents and "distant relatives" provided the most robust prediction accuracy across all simulations. Within-family accuracy was also lower in cross-generational selection. Interestingly, in models incorporating larger training sets with additional unrelated trees, we observed the opposite trend where within-family accuracy consistently improved.

Keynote Speakers

Mats Berlin
The Forestry Research Institute of Sweden

ASSISTED MIGRATION IN PRACTICE

Mats Berlin[1]

[1] Researcher, SkogForsk, The Forestry Research Institute of Sweden, Isafjordsgatan 22, 164 40 Kista, Sweden



Assisted migration involves using the intraspecific variation in adaptive traits to pro-actively transfer trees to better match the future climatic conditions. By sharing tree performance data across countries, we have developed joint Nordic assisted migration models (AMM) for Norway spruce and Scots pine and implemented them in a decision support framework, Planter's guide (PG) providing deployment recommendations for commercially available seed sources for operational forestry. There are two tools, PG and PG optimization, aiming to assist small forest owners or large forest owners and nurseries respectively. Both tools are freely available from the Skogforsk web page and have been designed in close collaboration with end users. This presentation will cover both the scientific background of the Nordic AMM and their implementation in the PG tools.

A CLIMATE-INFORMED SEED SELECTION TOOL FOR NORTH AMERICA: NA-SST

Nicholas Boyce [1], Genevieve Dorrell [1], Zach Zimmerman [1], and Andreas Hamann [1]

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As climate change is reshaping habitat conditions, ensuring the long-term health and productivity of forest ecosystems requires climate-informed seed transfer strategies. This may involve new species choices for reforestation, but more often can be addressed by matching planting stock from different, locally adapted source populations to new planting environments within the species range. The North America Seed Selection Tool (http://tinyurl.com/NA-SST) provides a framework to inform assisted migration and seed transfer across North America, covering all major tree species in the United States, Canada, and Mexico (the latter under development). NA-SST also visualizes historical and projected climate change for every potential target location to help evaluate which ecosystem types and species are likely to be supported in the long-term. The tool relies on forest inventory-based species distribution modeling for the historical 1951-1980 climate normal period to create a reference where species populations are assumed to be in equilibrium with climate conditions. Then, target ecosystem climates for the 1990s, 2020s, 2050s, 2080s are matched with historical climates of source ecosystems (1960s) using multivariate climatic distance matrices for 12 bioclimatic variables. The tool guides assisted migration in two steps: (1) species selection by identifying where a species' relative abundance is expected to remain stable, decline, or increase; (2) identifying seed source locations based on climate matching. For example a user can specify that the seed sources must be a reasonable climate match for both current (2020s) climate as well as still being a viable choice under anticipated climate change (e.g. 2050s).This presentation will showcase the tool's current capabilities and discuss planned extensions and improvements, including enhanced uncertainty quantification and inclusion of operational seed zones as selectable target regions, as well as seed collections orchard seed as potential sources to facilitate cross-border transfers of seed sources at the state/province and national level.

Keynote Speakers

Jessica Savage
University of Minnesota

CLIMATE CHANGE AND FALSE SPRINGS: WHEN MILD WINTERS BECOME DANGEROUS

Jessica A. Savage[1]

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As winter temperatures continue to warm, many temperate forests are experiencing early bud break and initiation of growth when there is still a high risk of freezing damage. As a result, some models have predicted that climate change will lead to an increase in the frequency of freezing damage as a result of "false springs". In many woody species, flowers are one of the earliest organs to emerge in the spring. Although floral freezing damage has been well studied in crop plants like apples, not much is known about the freezing tolerance of native flowers. Over the last five years, we have completed a set projects examining floral freezing tolerance, how to measure it, how it relates to flowering time, and the downstream impact of freezing on plant reproduction. We have found that species that currently bloom when the risk of freezing is low are the most vulnerable to false springs and that freezing damage to flowers can lead to decrease reproductive output and fruit development in woody plants. We are currently expanding on this work and collaborating with community scientists to better understand how climate change will impact plant phenology. In this project, we are examining intraspecific variation in the response of plants to spring warming and false springs. Our goal is to develop a more comprehensive view on the impact of spring warming on temperate forests in northeastern Minnesota.

THE ROLE OF CLIMATE AND INTERSPECIFIC GENE FLOW IN STOMATAL EVOLUTION ACROSS A P*OPULUS* HYBRID ZONE

Michelle Zavala-Paez [1], Stephen Keller [2], Jason Holliday [3], Matthew C. Fitzpatrick [4], and Jill Hamilton [1]

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Stomata play a critical role regulating plant responses to climate. Where sister species exhibit stomata trait differentiation, interspecific gene flow may impact the evolutionary trajectory of stomata trait variation with consequences to environmental adaptation. In this study, leveraging six latitudinally-distributed transects spanning the natural hybrid zone between *Populus trichocarpa–P. balsamifera*, we used whole genome resequencing and replicate common garden experiments to test the role interspecific gene flow and selection play to stomatal trait evolution. Geographic cline analyses suggested limited evidence for intrinsic barriers maintaining species-specific stomatal trait variation. Instead, differential selection associated with climatic gradients strongly influenced the distribution of genetic variation underlying stomatal traits. Using admixture mapping, we identified *SPEECHLESS* and *ABA DEFICIENT 1* as key loci associated with stomatal trait variation in hybrids. Consistent with genome-wide measures, climatic gradients as opposed to geographic gradients structural candidate loci associated with stomatal trait variation. This suggests that climate-driven selection, regardless of species identity, may be critical to stomatal trait evolution. These results point towards the critical role of climate in shaping the evolutionary trajectory of stomata in *Populus* and the value interspecific gene flow has to creating genetic recombinants needed for species to expand and persist across new environments.

Keynote Speakers

Alayna Mead
Pennsylvania State University

GENETIC VARIATION UNDERLIES DIFFERENTIAL RESPONSES TO CLIMATE CHANGE IN TREES: PREDICTING AND TAKING ADVANTAGE OF RANGEWIDE HETEROGENEITY

Alayna Mead[1]

[1] Postdoctoral Fellow, Department of Ecosystem Science and Management, Pennsylvania State University (alaynamead@psu.edu)



While novel climates are likely to pose a challenge to many species, particularly to long-lived and slow-evolving trees, natural populations often contain abundant genetic variation underlying fitness differences across climates. Tree populations are frequently locally adapted, having higher fitness in their local environment than non-local genotypes, so populations across a complex landscape likely vary in their tolerance to stressors such as high temperatures, drought, and other extreme events. Characterizing genetic variation and its effects on phenotypes will be necessary to predict fine-scale responses to climate change and identify management strategies that could benefit tree populations under future climates. I will discuss my work in two systems, oaks and poplars, in which I use genomic data to predict responses to climate change and inform conservation strategies while considering intraspecific genetic variation. First, in island oak (*Quercus tomentella*), a rare species present on six islands, I used genome-environment associations to estimate maladaptation to climate for local oak populations and under two forms of assisted gene flow. These results translated genomic information into risk metrics, enabling managers to compare outcomes of different pairs of seed sources and planting sites when considering restoration projects. Second, as part of the PopUp Poplars project, replicated clonal genotypes from the *Populus trichocarpa* and *P. balsamifera* hybrid zone were planted into 17 common gardens across the US. Using this dataset, I characterized reaction norms for each genotype and how they varied with species ancestry and site of origin. We predicted that genotypes with more *P. trichocarpa* ancestry may gain an advantage under warmer winter climates, highlighting that genotypic variation in responses to temperature could have landscape-level effects. In both studies, genetic variation is expected to influence responses to climate change, illustrating the importance of incorporating genetic data into climate change predictions for trees.

UTILIZING SOUTHERN OAK GENOTYPES AS A RESTORATION STRATEGY IN OUR NORTHEASTERN FORESTS

Samantha E. Fuentes-Gigliotti [1] and Myla F.J. Aronson [1]

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Regeneration failure of canopy trees due to overabundance of white-tailed deer and invasive non-native plant species is a critical issue defining the present and future of forests in urban and suburban landscapes. Compounding these pressures with climate change motivates land managers to identify strategies that can be implemented today while supporting future conditions and challenges. The objective of our study was to compare the use of local and southern genotypes (Kentucky) in canopy gap restoration. We conducted our study within Hutcheson Memorial Forest, an old-growth forest located in Somerset, NJ, where we planted native canopy trees (*Quercus alba* and *Quercus rubra*) within 10 canopy gaps (5 gaps planted with local genotypes and 5 gaps planted with southern genotypes). We used survival and height growth as success metrics and found that southern genotypes had greater success from 2021 to 2024. Southern *Q. alba* had a higher probability of surviving to the next year compared to local oak genotypes (year 1=3.3x, $p<0.01$; year 2=4.4x, $p<0.0001$; and year 3=3.0x, $p<0.01$). Southern *Q. rubra* also had higher probability of surviving to the next year (year 1=50.7x higher, $p<0.0001$; year 2=15.3x higher, $p<0.0001$). Growth data suggests similar results with highest growth occurring in southern oak genotypes particularly in years 1 and 2. Southern *Q. alba* had higher growth compared to local genotypes (year 1=11.2±10.4cm and 6.77±16.9cm, $Z=-2.555$, $p<0.05$; year 2=6.47±15.9cm and -1.04±17.0cm, $Z=-3.812$; $p<0.01$). Southern *Q. rubra* also showed higher growth than local genotypes (year 1=20.0±14.6cm and 3.78±10.8cm, $Z=-8.186$, $p<0.001$; year 2=8.10±21.cm and 0.502±5.76cm, $Z=-4.149$, $p<0.001$). Here our data suggests that restoration success is greater in southern oak genotypes compared to local oak genotypes. Future measurements will provide us with long term metrics on success that will support land managers looking for adaptive approaches of climate resiliency in restoration work.

Keynote Speakers

Stephen Keller
University of Vermont

ADDRESSING CONSERVATION PRIORITIES AND KNOWLEDGE GAPS WITH HELP FROM GENETICS – A STORY OF PARTNERSHIPS IN SUPPORT OF RED SPRUCE FORESTS

Stephen Keller [1]

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Forest trees are facing numerous threats under global change, including increasing climate stress and frequency of extreme events, but also legacies of demographic loss and range restrictions due to land-use change and development. Restoring fragmented forested landscapes and mitigating climate stress are challenges that conservation managers are facing in their efforts to build resilient, adaptable, and connected forest ecosystems. However, approaching these activities also presents complex issues for conservation management that often involve genetics: Should we mix seed lots for restoration or keep them separate? Is local still best, or should we move trees to keep up with climate change, and if so, from where, and where to? Are naturally occurring hybrids sources of variation that should be integrated into restoration programs or avoided? Forest geneticists have a fundamental role to play in helping to address these and other questions central to conservation practices and helping guide practitioners working to mitigate climate stress and maintain connectivity across the landscape. Here, I present highlights from the past 10 years of partnering with a dedicated network of conservation practitioners and forest managers working to restore red spruce (*Picea rubens* Sarg.) forests and foster the climate resiliency of this iconic conifer throughout its range across the Appalachian Mountains. While the questions are indeed complex, the partnerships have enabled a dialogue between geneticists and managers that has produced important new insights, opportunities, and pathways forward in the service of forest conservation.

Session 1A: FOREST HEALTH, PESTS, AND PATHOGENS

THE LINGERING HEMLOCK SEARCH PROJECT: WE NEED YOUR HELP TO RESTORE THE MIGHTY HEMLOCK

Olivia W. Hall [1]

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To date, eastern hemlocks have displayed little resistance to hemlock woolly adelgid (HWA) at the population level. The majority of eastern hemlock is susceptible to damage from HWA and therefore likely to decline and eventually die once infested with HWA. Breeding HWA- resistant hemlock populations of hemlock is a critical piece of an overall integrated pest management strategy to save eastern hemlock trees from the plight of HWA. In order for researchers to breed native HWA-resistant hemlocks, surviving, or "lingering," trees that may harbor a small amount of genetic resistance need to be found among wild populations. Then, these trees can be tested in a controlled setting and, if found to demonstrate some resistance, bred to increase the level and frequency of resistance. In order for researchers to breed a resistant hemlock, it is important to identify lingering hemlocks for additional evaluation and genetic research. This is where tools like the Lingering Hemlock Search protocol (developed as part of the Nature Conservancy's Tree Species in Peril initiative) and the TreeSnap application come in. We are sending out a call to anyone who is in the forest to be a part of this effort by helping us find and record these lingering hemlocks.

ADVANCING GEMONIC RESEARCH AND CONSERVATION EFFORTS FOR A NORTH AMERICAN WALNUT THREATENED BY A NON-NATIVE FUNGAL PATHOGEN

Cynthia N. Webster [1], Cristopher R. Guzman-Torres [1], Anthony He [1], Keertana Chagari [1], Amanda Mueller [1], Vidya Vuruputoor [1], Stefan Wnuk [1], Harshita Akella [1], Emily Strickland [1],Emily Trybulec [1], David Baukus [1], Laurel Humphrey [1], Owen McEwing [1], Michelle L. Neitzey [1], Nicole Pauloski [1], Emry Brannan [1] , Rachel J. O'Neill [1], and Jill L. Wegrzyn [1] ,Karl C. Fetter [2], Aziz Ebrahimi [3] , Martin Williams [4] ,Jeanne Romero-Severson [5], Sean Hoban [6], Keith Woeste [7], Carolyn C. Pike [7]

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Advancements in high-throughput sequencing and assembly technologies have greatly

Presentation Details

reduced the time and resources needed for genome construction, facilitating new ways of exploring how species and their populations have changed over time. *Juglans cinerea* (butternut) is a North American walnut tree threatened by non-native fungal pathogen, *Ophiognomonia clavigignenti-juglandacearum* (*Oc-j*). Discovered in the 1960s, *Oc-j* has devastated more than 80% of this North American species. Infected trees exhibit visible lesions and generally die within 5 to 10 years. While susceptibility to *Oc-j* is high in *J. cinerea*, its allopatric relative, *Juglans ailantifolia* (Japanese walnut), exhibits natural resistance. Interestingly, crossing both species yields a hybrid with intermediate resistance. As part of the Biodiversity and Conservation Genomics program at the University of Connecticut, the first reference genome for *J. cinerea* was assembled. While this 539 Mb chromosome-scale assembly from New Brunswick has provided a valuable framework for aligning sequencing data and conducting comparative analyses, resistance mechanisms are still poorly understood. To improve resolution, we assembled the first *J. ailantifolia* genome and an additional *J. cinerea* accession from Morton Arboretum in the United States. Our objective was to uncover genetic factors differentiating *J. cinerea* from *J. ailantifolia* in their response to *Oc-j* canker disease, and to understand how the US *J.cinerea* accession differed from the geographically isolated Canadian accession. To accomplish this, we conducted a gene family analysis across Juglandaceae classifying rapidly expanding and contracting gene families related to disease resistance. Furthermore, we built a graph pangenome composed of both *J. cinerea* individuals and the Asian walnuts (*J. ailantifolia* and *Juglans mandshurica*) to capture core, dispensable, and unique genes in a presence-absence variant-based analysis. While limited to only a handful of accessions, this research provides a genomic framework for future population-scale studies and informs conservation strategies for butternut.

STRUCTURAL VARIATION AND ASSEMBLY ARTIFACTS IN TWO *FAGUS GRANDIFOLIA* GENOMES INVESTIGATING CANDIDATE GENES FOR BEECH BARK DISEASE RESISTANCE

Airianna McGuire [1], Susan L. McEvoy [2], Adrian F. Powell [3] Susan R. Strickler [4], Carolina Jara [1], Cynthia N. Webster [1], Vidya S. Vuruputoor [1], U. Uzay Sezen [6], Nathan G. Swenson [6], Mohamed Elgallad [3], Fay-Wei Li [3], Eric J. Richards [3], and Jill L. Wegrzyn [1]

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American beech (*Fagus grandifolia*) is a keystone species in eastern North American forests associated with both carbon sequestration and nut production for wildlife. Across large portions of their expansive range, beech are facing widespread mortality from both Beech Bark Disease (BBD), caused by the scale insect *Cryptococcus fagisuga* and fungi *Neonectria ditissima* and *N. faginata*, as well as Beech Leaf Disease (BLD) caused by the *Litylenchus crenatae mccannii* nematode. While both diseases are more concentrated in the Northeast, they are rapidly spreading through the range. The focus of this work on BBD leverages the fact that 1–5% of trees exhibit natural resistance. Earlier work in the system, with transcriptome-derived markers, identified four SNPs linked to BBD resistance in a mapping population. To investigate resistance loci, two *Fagus grandifolia* reference chromosome-scale genomes were assembled: one from a resistant individual and another from an individual of unknown susceptibility. Hybrid genome assembly with MaSuRcA included deep coverage of both nanopore and Illumina short reads. Public Illumina RNA-Seq reads supported genome annotation. Moderate coverage sequencing of an additional five individuals was used to examine additional variants, including structural variation in the regions of interest. A candidate resistance gene (*metallothionein type 2)* was identified through alignments of the four markers previously associated with resistant individuals in a mapping population that segregated as a major gene.

ISOBORNYL ACETATE OVERDRIVE: AN 'OMICS' LENS INTO THE EASTERN HEMLOCK'S SEASONAL DEFENSE STRATEGY AGAINST THE HEMLOCK WOOLLY ADELGID

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The eastern hemlock (*Tsuga canadensis*), a foundational species spanning over 2.3 million acres in eastern North American (NA) forests, faces ecological collapse due to the invasive hemlock woolly adelgid (HWA; *Adelges tsugae*). First detected in 1951 and likely introduced via infested ornamental plants from East Asia, HWA has devastated NA hemlocks, which lack evolved defenses against this sap-feeding pest. In contrast,

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Asian hemlocks—including Chinese (*T. chinensis*), Ulleung (*T. ulleungensis*), and Siebold's (*T. sieboldii*) species exhibit natural tolerance shaped by co-evolution with HWA. NA hemlocks (*T. canadensis* and *T. caroliniana*), however, evolved defenses against native defoliators like the hemlock looper (*Lambdina fiscellaria*), leaving them vulnerable to the introduced pest, HWA. However, it was observed that interspecific variation in HWA response exists in eastern hemlock populations. A 2011 citizen science initiative identified the "Bulletproof" (BP) stand, NJ—this stand was hypothesized to be 'resistant' to the HWA, and clones of the BP stand were shown to retain a lower density of HWA in its canopy. This study investigates seasonal terpenoid and gene expression dynamics across five hemlock species (three Asian, two NA) and compares putative lingering (BP) versus susceptible *T. canadensis* individuals over a 12-month cycle across three sites. Key findings reveal *T. canadensis* produces the highest levels of isobornyl acetate among all species, and within eastern hemlock individuals, this monoterpene peaks in lingering populations during late fall and early winter. This seasonal surge coincides with feeding activity by HWA's cold-hardened sistens generation. To enable gene expression comparisons, we generated the first chromosome-scale genome assembly for *T. canadensis* (12 chromosomes, N50 = 1.4 Gb, 91% BUSCO completeness). The genome, dominated by LTR retrotransposons (58%) and encoding 35,810 annotated genes, provides a critical resource for comparative analyses of terpene synthase evolution and disease-resistance loci across gymnosperms.

TURNING INVASIVE PESTS AGAINST THEMSELVES – PROTECTING TREES WITH ANTI-PEST-SPECIFIC TREATMENTS

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Declines in the health and integrity of forests threaten forest product industries, ecosystem services, and regional economies. Advances in genomics are providing new opportunities for molecularly based forest pest management. One approach is to silence the essential genes required for the survival or function of forest pathogens or pests

using the RNA interference (RNAi) pathway. Unlike traditional pesticides, RNAi can be rapidly developed using the genome sequence of the targeted pest species. RNAi has shown to be effective against a variety of fungal pathogens and insect pests in agriculture and recently shown to be applicable against forest insect pests. The goal of our study is to develop and test RNAi-based suppression of six important test cases in forest trees including: chestnut blight, Dutch elm disease, butternut canker, laurel wilt, emerald ash borer and related invasive wood borers, and invasive adelgid species, including hemlock woolly adelgid. This presentation will provide an overview of our approach to developing RNAi as a tool in these systems, including identification of candidate genes for silencing, development of double stranded RNAs targeting candidate genes, uptake of double stranded RNAs by pathogen/insect pests, and in vitro and in planta evaluation of efficacy. Initial results will be presented for chestnut blight and emerald ash borer.

AN ASSAY TO ACCELERATE SCREENING FOR RESISTANCE TO DUTCH ELM DISEASE IN AMERICAN ELM TREES

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The Northern Research Station, in collaboration with many federal, state, university and non-profit partners, leads an effort to screen large survivor American elms for resistance to the non-native pathogens responsible for Dutch elm disease (DED); *Ophiostoma ulmi* and *O. novo-ulmi*, for use in genetically diverse American elm seed orchards. Field phenotyping is critical to evaluate the "real world" disease resistance and performance of survivor elms. It is, however, a lengthy process that requires large plots of land and intensive management. Greenhouse assays using young potted trees can be useful to screen out individuals with low levels of resistance, increasing the efficiency of field testing. The main goal of this study was to develop a practical and reliable potted tree screening assay to evaluate resistance of survivor American elm clones and progeny to DED. Over the course of three years (2021-2023), we evaluated the effects of (i) dose response, (ii) tree age and (iii) inoculation methods on disease symptom development in American elm clones of known susceptibility. In 2024 we used the findings of these tests to trial a refined approach to evaluate DED resistance of both ramets and progeny of large survivor elms from our American elm breeding program. The results demonstrate that disease response was independent of *O. novo-ulmi* dose within the range we tested (2500 – 25,000 spores/tree). Older elms generally produce stronger symptoms, however

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segregation among genotypes of known levels of resistance was evident even in 1-year-old trees. A hanging drop method of delivering inoculum to elms produced similar symptom severity to a traditional scalpel inoculation, while reducing wounding. Finally, foliar decline from the potted assay was highly correlated with field canopy decline for individuals of the same genotypes. These results suggest a greenhouse assay can reliably screen survivor elms for DED resistance, facilitating increased efficiency of field testing.

Session 1B: TREE IMPROVEMENT AND BREEDING

COLLABORATIVE SEED ORCHARD PROGRAM TO SUPPLEMENT WHITE OAK (*QUERCUS ALBA*) TREE IMPROVEMENT EFFORTS

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The high degree of genetic variability in white oak (*Quercus alba*) provides significant opportunities for tree improvement. The White Oak Genetics and Tree Improvement Program (WOGTIP) is a collaborative effort to quantify white oak genetic variation, improve traits that will promote seedling survival and growth in natural forests, and improve traits that have economic and ecological value in this keystone species. WOGTIP supports forest management efforts through creation of a sustainable supply of genetically improved, high-quality seedlings to supplement advanced regeneration and for reforestation. Progeny tests have been established throughout white oak's geographic range using regionally based seed sources, and there is a range-wide provenance test. The progeny tests will be converted to seed orchards, while the range-wide test will be maintained into perpetuity for research. Parents of superior progeny will be used to create grafted seed orchards. Despite these efforts, demand for acorns that produce high quality seedlings will significantly exceed what WOGTIP can supply to the nurseries. A novel program is being piloted in KY to address this problem. The Independent Seed Orchard Program (ISOP) is a collaboration between UK Forestry Extension, KY Division of Forestry (KDF), and private landowners and other partner participants with a goal of creating small (0.5 acre) seed orchards across Kentucky with 10-15 landowners being enrolled each year. ISOP sites are approved by a KDF professional forester, and the landowner is provided 100 of the top 1% of seedlings from the KDF nursery. Seed orchard sites are "certified" as part of WOGTIP and acorns produced will go back to the nursery for production of superior seedlings. Landowners get advice and guidance throughout the process from the KDF professional foresters and UK Forestry Extension. It is hoped the ISOP concept described in this presentation will be adopted by other states.

Presentation Details

EFFECTS OF ABSCISIC ACID ON MATURATION AND GERMINATION OF NORTH AMERICAN ASH (*FRAXINUS* SPP.) SOMATIC EMBRYOS

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Since 2002, the massive loss of ash trees to the invasive insect pest emerald ash borer (EAB; *Agrilus planipennis*) has drastically altered North American forests east of the Rocky Mountains and has devastated specialized forest ecosystems and urban landscapes. Due to this infestation, species predominantly found in the eastern United States such as green ash (*Fraxinus pennsylvanica*) and white ash (*Fraxinus americana*) have been largely extirpated from their native ranges. EAB was discovered for the first time west of the Rockies in 2022, where it now threatens Oregon ash (*Fraxinus latifolia*) with functional extinction. Clonal propagation of this species and other *Fraxinus* species would be a valuable tool for germplasm conservation and eventual restoration. In vitro clonal propagation techniques such as somatic embryogenesis (SE), offer a powerful approach to produce clonal ash planting stock. The optimization of SE in North American ash is critical, as the EAB population continues to spread and inflict devastation onto additional ecologically significant species, like Oregon ash. One avenue for improving SE productivity is the use of plant growth regulators, such as abscisic acid (ABA), to enhance somatic embryo development and maturation. Previous studies have shown that low doses of ABA can improve somatic embryo development in several crop and forest species. To investigate this relationship in North American ash, we conducted several experiments to optimize SE protocols, including: (1) varying ABA exposure durations in liquid suspension culture, (2) testing different ABA concentrations in semisolid media, and (3) testing combined treatments of ABA and cold pre-germination treatment durations. Our preliminary results align with prior findings in other tree species, supporting a significant role of ABA in the enhancement of somatic embryo development and maturation. These results will refine SE protocols for ash, accelerating reforestation efforts for Oregon ash and *Fraxinus* species threatened by EAB.

PERFORMANCE OF TOLERANT POPULATION OF EUROPEAN ASH IN CLONAL AND PROGENY TRIALS IN SWEDEN

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European ash (*Fraxinus excelsior*) is a keystone tree species whose future is threatened by the invasive pathogen *Hymenoscyphus fraxineus*, the causal agent of ash dieback, which has led to an extensive decline of ash populations across Europe. Consequently, the loss of ash has led to habitat degradation for numerous organisms relying on ash as

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a substrate. Breeding for resistance is the most feasible measure to ensure ash conservation as the presence of resistance has been demonstrated in several genetic trials in Europe. In Sweden, efforts have been made to develop a resistant ash population. Between 2016 and 2022, four clonal trials were established, involving over 200 clones. These genotypes were previously selected based on extensive surveys conducted between the years 2013 and 2019 in forests, natural landscape, and seed stands throughout the natural distribution range of ash in the southern Sweden. In addition, a progeny trial was established in 2014 with seedlings derived from seeds of 13 tolerant and susceptible clones from an open-pollinated seed orchard. To evaluate ash dieback tolerance, annual health assessments were conducted in all trials, ensuring continuous monitoring of disease progression and tree vitality. Here, we present the results of a quantitative genetic analysis and provide a descriptive overview of the experiments. The findings demonstrate the feasibility of obtaining tolerant ash populations in Sweden, marking significant progress in breeding for ash dieback resistance.

A COST-EFFECTIVE APPROACH TO IMPLEMENTING SEED SOURCE TRIALS IN THE PACIFIC NORTHWEST

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In the Pacific Northwest, assisted population migration (APM) has been discussed as a solution to realign the optimal match between tree seed source and planting location during reforestation. Previous studies on APM have, however, included a relatively low number of testing sites that run long-term. Our group at Washington state Department of Natural Resources (WA DNR) has begun work to establish a large network of seed source trials across various land ownerships (small private, public, etc.) with the widespread and commercially valuable coastal Douglas-fir (*Pseudotsuga menziesii var. menziesii)*. Our primary goals are 1) to establish numerous trials rapidly, generating more data to bolster the Douglas-fir climate transfer function, and 2) to provide demonstration examples that allow forest managers to observe the performance of different seed sources in their local area, stimulating the conversation around APM. We have established 22 trials since 2022 (2-CA, 7-OR, 12-WA, 1- BC) using a relatively simple trial design that enables easy and inexpensive installation and requires minimal maintenance to accommodate landowners with limited resources, especially small private landowners. Twenty-three seed sources have been selected for this project– 18 are “woods-run” seed sources (collected from natural stands out in the woods) with six each from Washington, Oregon, and California that represent a wide climate range within each state and an even distribution geographically. The remaining five sources are orchard lots produced at WA DNR’s seed orchard, which represent the status quo in this project. Preliminary data from the 2- and 3-year-old trials will be presented and anecdotes on the successes and challenges of our cross-sector implementation approach will be discussed as an update to our ongoing project.

INCORPORATING GENETIC GAINS INTO LOBLOLLY PINE YIELD MODELING

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Incorporating genetic gains into growth and yield modeling is crucial for optimizing forest management planning. This talk will begin with a review of studies on this topic focusing on loblolly pine in the southern United States. A case study on the topic in the western Gulf region will then be presented. In the western Gulf region, two models, one for loblolly pine plantations established in the late 1970s (wood-run model) and the other for intensively managed (including planting improved seed lots) loblolly pine plantations established in the 2000s are available. Long-term progeny trial data are fitted to both models and their differences were compared and discussed in terms of integrating genetic gains into growth and yield modeling.

AN INTEGRATED APPROACH TO UNDERSTANDING ADVENTURE ROOT INDUCTION IN RECALCITRANT SPECIES

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Cutting propagation provides uniform plant materials that retain the original characteristics of the donor. Additionally, plants generated by cutting reach the reproductive phase sooner because they bypass the immature seedling phase. This can save time and money for commercial plant production. Thus, propagation by cutting plays a significant role in horticulture, agriculture, and forestry. However, this method requires the formation of adventurous roots (ARs), and substantial variation in the ability to form ARs exists among different species and cultivars within the same species. By comparing recalcitrant species, chestnut (Castanea) and yellow camellia (YC) (*Camellia nitidissima*), to easy-to-root poplar (*Populus*) and weeping willow (*Salix babylonica*), it is found that the semi-lignified stem cuttings of American chestnut (AC) and yellow camellia (YC) exhibit low levels of rooting-promoting hormone indole-3-acetic acid and high levels of other hormones that inhibit rooting induction and development. Additionally, their hormone distribution between leaves and stems differs from that of easy-to-root poplar. Profiling of primary and secondary metabolites indicates that AC and YC contain fewer AR-promoting compounds and more AR-inhibiting ones. Thus, both AC and YC have unfavorite endogenous hormone and metabolite profiles that may contribute to their recalcitrance to rooting. Our study lays the foundation for future investigation of genes and gene networks that regulate AR induction in recalcitrant species. Furthermore, AC stems possess a closed sclerenchyma ring, while the sclerenchymata in YC, poplar, and weeping willow cuttings are dispersed. A closed sclerenchyma ring may hinder AR emergence, as AR primordia must force apart the sclerenchyma before roots can outgrow. Results of promoter and RNA-seq analyses will also be presented at the meeting.

Presentation Details

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Session 1C: POPULATION AND QUANTITATIVE GENETICS

HYBRIDIZATION AND ADAPTATION: THE GENETIC HISTORY OF SHORTLEAF PINE

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Shortleaf pine (*Pinus echinata* Mill.) is an ecologically and economically important conifer with a broad distribution across the southeastern United States. Shortleaf pine has a history of hybridization and introgression with loblolly pine (*Pinus taeda* L.) and pitch pine (*Pinus rigida* Mill.) which may have been driven by ecological and climatic factors. We used a 50k SNP chip to analyze 241 pine samples, including 224 shortleaf pines from across the species' range, to examine population structure and genetic variation. Samples from the Mount Ida seed orchard (Mt. Ida, AR), the Shortleaf Pine Seed Source Study (Crossett, AR), and wild collections were used to represent these populations. Three main clusters were identified using sNMF and DAPC analyses which corresponded to geographic location and presence of hybrid ancestry. A number of shortleaf trees exhibited a small proportion of SNPs (3-12%) of loblolly origin suggesting ancestral loblolly hybridization despite a shortleaf phenotype. Evidence of ancestral loblolly hybridization (admixture) was found in all individuals of the Oklahoma and northwest Arkansas (Ouachita and Ozark Mountains) populations (all allopatric with loblolly pine), while no admixed individuals were identified from the Coastal Plain and Piedmont populations (all sympatric with loblolly pine). Populations from allopatric regions outside of southeast Oklahoma and northwest Arkansas contained a mix of individuals, those with and without hybrid ancestry. These populations exhibited high g^2 consistent with population stratification. We hypothesize that edaphic factors and pathogen resistance may be responsible for the persistence of this hybrid signal in allopatric portions of the shortleaf pine native range. The results of this study will be used to inform shortleaf pine breeding and conservation efforts.

Presentation Details

UNRAVELING POPULATION STRUCTURE AND GENETIC DIVERSITY OF EASTERN COTTONWOOD FROM THE U.S. FOREST SERVICE BREEDING PROGRAM

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Eastern cottonwood (*Populus deltoides*) is a model platform for carbon fixation and storage, phytoremediation, water and nutrient use efficiencies, biofuel quality, and urban site tolerance. Over time, the US Forest Service, in their Southern Research Station, has created a highly valuable clone collection from all Southeast states. These clones have shown high phenotypic performance increasing national and international demand for *P. deltoides* clones in the last decade, primarily driven by carbon sequestration applications for climate mitigation. Advances in molecular techniques make it possible to understand valuable information, such as fingerprinting genetic structure and diversity. This project aims to identify the population structure and produce and archive the molecular identities of *P. deltoides* Stoneville clones and their genetic diversity. Leaves were collected from 300 clones in the original stool bed and breeding orchard in Stoneville, Mississippi. Genotyping from DNA extractions was performed using a probe SNP (Single Nucleotide Polymorphism) chip array. After quality cleaning and variant calling, 21,000 of the most informative SNPs were selected. Preliminary results from the first 95 accessions show six subpopulations based on their degree of relatedness with considerable genetic variance. The structure population was able to identify the origin of some unknown clones. The following steps are: (1) Include genotyping data from the remaining 205 clones, (2) Upload the genetic identity of each clone to an open-source repository for germplasm conservation purposes. Our results will provide critical first steps to ecological genetics for restoration with *P. deltoides* under USDA Forest Service programs and private NGO programs.

CHANCE OR PATTERN: DISTRIBUTION OF MAJOR GENE RESISTANCE TO THE NON-NATIVE PATHOGEN THAT CAUSES WHITE PINE BLISTER RUST

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While tree host traits that confer resistance to new diseases are adaptive in the presence of an introduced pathogen, the origins of these traits and their selective value before pathogen invasion (i.e., in naïve populations) are largely unknown. Consequently, our ability to predict the geographic distribution of resistance traits in forest pathosystems that lack a coevolutionary history is limited, as is our ability to predict the outcome of selection by a new pathogen on a host's fundamental niche. Exploring trait correlations with abiotic and biotic factors can inform hypotheses to explain the distribution and frequency of a resistance gene against a non-native pathogen. Climatic variation associated with landscape patterns of resistance frequencies can provide insights and associations of stress tolerance traits with disease resistance can suggest possible shared functions. Then again, the resistance trait may be a relict that protected the host from a pest or pathogen that it was historically exposed, and the distribution of the trait reflects the proximity to that exposure. Alternatively, the resistance trait may be a product of a mutation with no constitutive cost or benefit to the host, yet that population served as the source for founding events across the landscape. The origin and distribution of a resistance trait may be a product of the interaction of factors. We will discuss these hypotheses and analyses to decipher patterns of major gene resistance to the non-native disease white pine blister rust in limber pine in western North America.

ENVIRONMENT-SPECIFIC GENOMIC PREDICTION OF ASH DIEBACK RESISTANCE IN EUROPEAN ASH (*FRAXINUS EXCELSIOR L.*)

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The invasive pathogen *Hymenoscyphus fraxineus* (HF) poses a severe threat to European ash (*Fraxinus excelsior* L.) in a changing climate. While genomic studies provide valuable insights into HF resistance, the complexity of high-dimensional genetic and environmental variables presents challenges in modeling genotype-by-environment (G×E) interactions. In this study, we analyzed whole-genome sequencing data (>11 million SNPs) from 320 ash trees across Poland. A genome-wide association study identified 120 SNPs associated with HF resistance, with over 40% located on chromosome 14. Genomic prediction using subsets of at least 2,500 SNPs yielded more than 80% accuracy in predicting tree health status. Incorporating environmental variables, including mean diurnal range (BIO2), temperature seasonality (BIO4), maximum temperature of the warmest month (BIO5), temperature annual range (BIO7), and precipitation seasonality (BIO15), improved the prediction accuracy by 3% to 8%. Cross-validation demonstrated that models trained and tested under similar environmental conditions benefited most from integrating G×E effects, whereas improvements were more modest in untested environments. These findings highlight the importance of combining genomic and environmental data to enhance resistance strategies for European ash. They provide practical guidance for targeted breeding and sustainable forest management, promoting the resilience of *F. excelsior* against ash dieback under diverse environmental conditions.

Presentation Details

SHEDDING LIGHT ON THE GENETIC ARCHITECTURE OF ADAPTIVE INTROGRESSION IN A POPULUS HYBRID ZONE

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Plant evolution in response to global change relies on standing genetic variation, the raw material upon which natural selection may act. However, hybridization and introgression may offer an additional source of recombinant variation to facilitate rapid evolution in response to changing selective pressures. Despite this understanding, the mechanisms through which introgression may influence adaptive potential in rapidly changing environments remains unclear. In temperate and boreal trees, the occurrence of hybridization during secondary contact has been promoted by climatic fluctuations during glacial and interglacial periods. *Populus trichocarpa*, a temperate forest tree species distributed across the Pacific Northwest of the United States and Canada hybridizes freely with *P. balsamifera*, a boreal species where the distribution range of the two species overlap. The resultant hybrid zones provide genetic variation upon which selection may act, and introgression may facilitate adaptation to novel environmental conditions beyond that of the parental species. Using whole genome resequencing data of parental and admixed genotypes of *P. trichocarpa* and *P. balsamifera* sampled from multiple replicated contact zones, we test for signatures of selection that might indicate adaptive introgression. To do this we will use selective sweeps based on estimates of genetic differentiation (Fst, Dxy) with ABBA BABA tests and Bayesian genome cline analysis. To tease apart the impact of selection from neutral demographic processes we will also quantify nucleotide diversity (π_W) across the genome and assess the distortion of site frequency spectrum at regions that are candidates for selective sweeps. To further identify the mechanisms contributing to adaptive introgression, we will test for the relationship between environment and introgression using latent factor mixed model. Given the repeated contact zones spanning environmental gradients between the two species, we expect to identify both genome wide signatures of adaptative introgression alongside transect-specific signatures that may inform the evolutionary trajectory of parents and hybrids under changing environmental conditions. Ultimately, the study will shed light on the role of hybridization in adaptation to climate change.

CAN ALLELE FREQUENCY DIFFERENCES BETWEEN POPULATIONS OF LOBLOLLY PINES BE USED TO SELECT FOR DROUGHT TOLERANCE?

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The commercial loblolly pine (*Pinus taeda* L.) forests of Texas are part of the Piney Woods (PW) of East Texas and Western Louisiana. Drought can be a problem with stand establishment and the summer dryness index is expected to increase. The Lost Pines (LP) are isolated populations of loblolly pine in central Texas and are more drought tolerant than trees in the PW. An understanding of the genes and variants that make the LP more drought-tolerant may help with the breeding of trees in the PW. We genotyped PW and LP trees as well as a progeny test of PW parents for 33,368 single nucleotide polymorphisms (SNPs). We compared allele frequencies to identify those SNPs with the greatest difference between the LP and PW. We chose 75 unlinked SNPs with frequency differences greater than 0.4 or greater than 0.3 but in genes previously found to be differentially expressed in response to drought stress. Within 49 PW maternal parents, we found large differences in the average frequencies of the "LP alleles," which ranged from 0.14 to 0.51. We examined progeny to see if maternal parents with allele frequencies more like LP trees have offspring that differ from those that are less similar. We found no relationship between δC13, an estimate of efficiency, and allele frequency, although we did find a strong relationship between δC13 and growth traits. We did observe a slight but significant positive correlation between parental LP allele frequency and survival and a significant negative correlation with both diameter and height. Mortality was not always caused by drought and was not correlated between the three progeny tests. However, severe mortality in four 2023 progeny tests has provided an opportunity to determine if frequencies of these 75 SNPs or others are correlated with survival.

Session 2A: CLIMATE CHANGE AND ADAPTATION

PINE SPECIES ALLOCATION ACROSS MULTIPLE COUNTRIES UNDER DIFFERENT CLIMATE SCENARIOS

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Forest plantations provide vital resources and products for all society. Climate-change scenarios expose forests to environmental hazards, resulting in tree mortality or decreased productivity. Recommending the right species to be planted in the right place is essential and tree breeding programs offer valuable data on species adaptability through field trials. However, even with this information, there is a lack of studies that can provide decision-support models capable of predicting the impact of climate change in site-species recommendations over different environments. This study aims to develop multi-country decision-support models for pine species that can assist in genetic material allocation under current and future climate scenarios. The dataset was composed of 553 genetic trials covering 12 different species of pine, including hybrids, distributed across 12 different countries. The variable selected to express growth potential was the dominant height at age 8 years (HT8). Environmental covariates were obtained from

WorldClim 2.1. Random Forest models were fitted for each genetic material and were used to predict HT8 values, generating maps of optimal species allocation. Model evaluation metrics were performed using R-squared (R^2); Root Mean Square Error (RMSE); Mean Absolute Error (MAE). The RF models showed high accuracy, with a mean R^2 of 0.78, MAE of 6.4%, and RMSE of 8.6% across all species. The most widely allocated pure species across both scenarios were *Pinus maximinoi*, *Pinus tecunumanii* high elevation, and *Pinus tecunumanii* low elevation, covering 28%, 16.9%, and 4.2% of the total area, respectively. Under the future scenario, the ranking of species remained consistent, though the proportions shift slightly. The proposed methodology delivers practical models to assist companies to select the best Pine species suitable to their sites. Further research will refine the models by adding tree breeding trial and inventory data from companies and incorporating more environmental variables from other sources.

ENVIRONMENTAL DRIVERS OF PLANTED SEEDLING MORTALITY IN THE PACIFIC NORTHWEST

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Forest trees face increased mortality due to climate change. Intensifying drought can lead to increased heat stress and wildfire prevalence, whereas extended warming periods can lead to increased prevalence of pests, pathogens, and invasive plants. Regeneration of natural and planted stands may be particularly affected by these challenges. Results of a detailed inventory of 24 northern states found nearly two-thirds of plots surveyed could face regeneration difficulties, while another study in the southwestern United States found planted conifer seedling survival rates averaged just 25% during post-fire regeneration. In this study, we aimed to identify the environmental drivers of planted conifer seedling mortality in the Pacific Northwest (PNW) of the United States. We acquired seedling survival data from Manulife Investment Management plots from across the PNW for 12 species planted between 2013 and 2022. Survival was assessed one and two years post-planting. Using a machine learning approach to assess the importance of climate, terrain, and ecological zone (EPA Level IV ecoregions and biogeoclimatic (BEC) zones), we found that, across the study area, ecological zones, species, and average climate in year of planting are highly important predictors of seedling survival. By implementing our model at a local scale, we identified spatially important environmental predictors for each species and generated spatial predictions for survival based on plot location and ecological zone. These predictions will help guide future seedling planting efforts while adjusting for future climate scenarios.

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INTROGRESSION AS A BUFFER AGAINST MALADAPTATION IN A KEYSTONE NORTHERN FOREST TREE

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Climate change is outpacing the rate at which forest trees can shift their ranges, leading to increasing maladaptation. The ability of tree species to persist will largely be determined by the standing genetic variation available in the population. The keystone northern forest tree red spruce (*Picea rubens*) is severely threatened by climate change due to low genetic diversity. However, it hybridizes with its sister species, black spruce (*P. mariana*), and most red spruce trees found in the sympatric zone derive significant portions of their genome from past hybridization with black spruce. Common garden studies suggest introgression from black spruce plays a key role in supporting red spruce's capacity to adapt to climate variation across its range, but much remains unknown about the spatiotemporal distribution of past hybridization, opportunities for secondary contact, and whether hybridization is ongoing in the sympatric zone. While species distribution modeling methods have been developed to incorporate intraspecific genetic structure, there is a notable lack of methods available for modeling hybrid zones and evaluating whether climatic or landscape factors facilitate or inhibit hybridization. Here, I present an array of possibilities for modeling the environmental niche of a hybrid system, including ancestry distribution models and occurrence models weighted by admixture. I jointly model the climate niche of red spruce and black spruce and project this model through time to evaluate changes in hybrid dynamics since the Last Glacial Maximum. I also project the model into the future to determine how black and red spruce's ranges are expected to shift relative to each other and whether we might see increased or reduced potential for hybridization. This work will enhance our understanding of hybridization as a mechanism to increase adaptive capacity and guide ongoing efforts to apply genomics to red spruce conservation and restoration.

FROM SEED TO SEEDLING: ADAPTATION TO CLIMATE IN EARLY LIFE STAGES OF *QUERCUS ALBA* POPULATIONS WITHIN AN ASSISTED MIGRATION FRAMEWORK

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Germination and seedling establishment are vulnerable and critical stages in the life cycle of trees. Even more so within an assisted population migration framework, which proposes the translocation of seeds from a warmer climate (provenance) to a colder plating site expected to warm during the lifespan of the trees. Selecting the right seed provenance (population), however, requires a balance between a population's ability to germinate and survive the early seedling stages under a climate colder than its provenance and optimizing growth under warmer conditions later in the tree's life. Understanding intraspecific adaptation to climate in early life-stages, and the effects of translocating warmer provenances to colder planting sites is then a must. We focus on *Quercus alba*, a wide-ranging species of ecological and economic importance. Using common gardens, we evaluate 17 populations from provenances representing a gradient of climates between 0.5 ºC to 8.5 ºC warmer than that of the common garden. Ten populations were planted in one common garden in the Fall of 2023, and seven populations were planted in two common gardens in the Fall of 2024. Traits observed included seed weight, germination rates, seedling height obtained three times during the growing season, and leaf senescence. Preliminary analysis detected clines in populations' average seed weight, with populations from warmest provenances having heavier acorns, and clines in populations' average seedling height, with populations from the warmest climates taller than those from the warmer climates, with both the steepness and the variation explained increased during the growing season. This was surprising because larger acorns characterizing the warmest populations were expected to have a larger effect in growth at the beginning of the growing season. Clines in fall phenology indicated that the warmest populations underwent senescence later in the season. Final analyses will inform best translocation practices in *Quercus alba*.

INTERACTIVE EFFECTS OF GENETICS AND ENVIRONMENT ON DOUGLAS-FIR SEEDLING RESPONSES TO DROUGHT AND EXTREME TEMPERATURE CONDITIONS

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Improving our understanding of genetic and environmental interactions can help guide landowners to more effectively deploy seedlings of different genetics across their ownership to optimize seedling performance and facilitate adaptation to the predicted changes in future climate conditions. This is particularly relevant for Douglas-fir (DF), a critical species in the timber industry of the United States' Pacific Northwest that faces warmer and drier growing season conditions. In this study, developed by the Vegetation Management Research Cooperative (VMRC) at Oregon State University, we are conducting a variety of physiological assessments for 55 unique DF genotypes

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(including 50 full-sib families of Coastal DF, 1 family of Interior DF and 4 hybrids of Coastal and Interior DF) to explore seedlings responses to a variety of environmental conditions with a focus on drought, humidity and temperature. These assessments include stomatal conductance sensitivity to vapor pressure deficit, xylem hydraulic conductivity and vulnerability to cavitation, frost hardening, and heat tolerance to identify drought-resistant and frost-tolerant genotypes that could be strategically deployed to mitigate the impacts of water stress and extreme temperatures in forest ecosystems. The goals of these assessments are to determine: 1) if there is strong genetic control in seedling responses; 2) which environment conditions associated with parent trees are driving certain responses; and 3) whether there are potential trade-offs between traits. Early results have demonstrated large variability in responses across genotypes to all physiological assessment conducted, demonstrating that there is strong genetic control. This study corresponds to Phase 1 of the Genetic x Environment x Early-Silviculture Interactions Study (GEnESIS), where a subset of genotypes selected in this phase will be tested under contrasting field conditions and silvicultural management treatments in Phase 2 of the study. Our results will be included in a decision support system that is currently being developed by the VMRC.

Session 2B: TREE IMPROVEMENT AND BREEDING

COMPREHENSIVE STRATEGIES FOR MANAGING GENE DIVERSITY AND GROWTH POTENTIAL OF CROPS BY MEAN OF SELECTIVE CONE HARVEST IN A SEED ORCHARD OF *PINUS KORAIENSIS*

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This study investigated the variations of cone production and general combining ability (representing growth characteristics) in a clonal seed orchard of *Pinus koraiensis* in South Korea. It estimated the impact of annual production variability on genetic gain and gene diversity and proposed comprehensive strategies for enhancing stable cone production and sustainable forest productivity. Utilizing cone and seed yield data over three years (2020–2022) from the clonal seed orchard, we estimated the effective population size and the gene diversity of seed crops. Results indicated significant annual and clonal variation in cone production, with a notable impact on the effective population size and the gene diversity. The study also categorized clones into four groups based on the coefficient of variation (CV) and clonal mean for cone production, examining the implications for genetic diversity and gains. By harvesting cones from the group with high yields and low CVs (variance), it is possible to minimize the reduction in genetic diversity caused by selection, as well as improve yield and growth potential of crops. In other words, regulating the CV may effectively reduce the loss of gene diversity and facilitate the re- selection of genotypes that consistently yield elite seeds with potentially superior growth traits. Our findings will contribute to the understanding of the dynamics of seed production, offering insights into the selection and management of clones for improved seed yield and maintain genetic diversity in seed orchards.

Presentation Details

POLLEN VIABILITY TESTING METHODS TO PREDICT SEED YIELDS IN LOBLOLLY PINE

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Loblolly pine pollen viability was tested using the industry standard in-vitro agar germination test while also taking this method one-step further to measure pollen grain size. Additionally, respiration testing using a new machine to measure rate of oxygen consumption (Hansatech Oxytherm+Respiration) and a new approach to testing loblolly pine pollen using impedence flow cytometry (Amphasys Ampha™Z30) was used to test loblolly pine pollen viability. Prior to testing, these pollen lots were applied to the branches of seven different orchard clones in the spring of 2020 using controlled-pollination techniques and cones were harvested from these crosses in the fall of 2021 to complete cone analysis and determine each pollen's ability to produce filled seed per cone. Impedence flow cytometry was found to have the strongest relationship in predicting filled seed per cone with an R^2 of 0.85, followed closely by in-vitro germination testing R^2 of 0.82 and then respiration testing with an R^2 of 0.66. Pollen lots having the lowest in-vitro germination rates were shown having distinct non-viable populations when tested using impedence flow cytometry and viewed in the AmphaSoft2.0 software. An additional 21 pollens were tested using these three testing methods and compared with each other but not taken to seed yield.

GIRDLING TOPGRAFTED *PINUS TAEDA* TO INDUCE STROBILI PRODUCTION

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Reducing the length of the breeding cycle is paramount for increasing the rate of genetic gain. Tree breeding and testing strategies have reduced the age of selection in every cycle. Inducing strobili production of young selections by top grafting their scion into reproductively mature rootstocks has become standard practice. Girdling is another technique to induce flowering but has not been routinely used in southern pines. In this study, we evaluated the use of three gridling techniques, overlapping trunk girdles and individual branch gridles using wire and sawing, in trees grafted by the Cooperative Tree Improvement Program at the Georgia Forestry Commission's Arrowhead Breeding Center. The girdling treatments were applied in June (when female strobili initiation occurs) of 2024. Forty top grafted orchard trees were assigned as either a control or treated with overlapping trunk girdles. Another 14 top grafted orchard trees had individual branches assigned to treatments as either a saw girdle, wire girdle, or control. Whole-tree strobili

counts and individual branch strobili counts were assessed in February 2025. Preliminary analyses indicated that the 2025 season was particularly prolific and the mean strobili per branch was high among all treatments, even the control. There was some indication that the probability of any strobili production was increased with girdling treatments for grafts made the previous year (one year after grafting). Additional measurements will be recorded during the 2025 breeding season, as well as in subsequent years. In Spring of 2025, an additional study was installed to evaluate flower stimulation treatments (girdling and gibberellin stem injections) in very young scion material (2-year from seed) compared to the conventional selection age (5-year from seed). Our findings are particularly important for the application of genomic selection, which permits selection of very young trees.

PHENOLOGY OF FLOWER RECEPTIVITY IN A *PINUS TAEDA* BREEDING ORCHARD CAN BE PREDICTED WITH HEAT SUMS

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Understanding and predicting the phenological stages of *Pinus taeda* flowering is crucial for optimizing controlled pollination and improving seed yield in breeding programs. This study evaluated heat-accumulation models for predicting the day of peak receptivity of female strobili for pollination using data from 2011 to 2024 at the Arrowhead Breeding Center near Cochran, GA, USA. We evaluated two different models (heat sums and growing degree days) using several combinations of base temperatures and start dates. Our findings indicate that the heat sum model with a base temperature of 50°F and a start date of January 1st provided the most consistent and accurate predictions with a critical threshold of 918 heat credits. The simpler model had an average error of 3.8 days at the Arrowhead Breeding Center, while it varied by 5.3 days at multiple locations. The clones used in the study also varied considerably in their day of flowering, with a range of 15 days between the earliest and latest clones. Based on the prediction model, we found that the date for bagging female strobili is safe when around 540 heat sums are accumulated. This study supports the use of heat sum models for phenological predictions, and we envision a "flowering countdown clock" tool to guide breeders as they plan their controlled pollination activities.

ROTATION-AGE ASSESSMENT OF REALIZED GENETIC GAIN IN SLASH PINE (*PINUS ELLIOTTI VAR. ELLIOTTI)*

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Forest tree breeding programs aim to maximize accumulation of genetic gain per unit time. However, genetic gain is typically assessed well before the primary phenotypes, volume and form of a stand of trees at rotation, can be measured. To maintain a reasonable rate of genetic gain and test a large population, tree breeders establish tests with single-tree plots and measure them at an early age. In addition to opportunity cost from not taking advantage of sexual maturity prior to rotation, integrity of the experiment design cannot be maintained over the long term. Furthermore, testing large populations makes block plots meant to resemble samples of single-family stands untenable because of land use, cost, time, and effort. There thus exists a question of (1) the extent to which genetic gain at an early age corresponds to genetic gain at rotation and (2) how genetic gain in the now-typical single-tree plots compared to that in stands of trees with one or two shared parents. The Cooperative Forest Genetics Research Program at the University of Florida established a series of multi-environment trials through the 1990s-2000s to answer these questions for a breeding population of slash pine (*Pinus elliottii* var. *elliottii* Englem.). Here we will discuss the genetic gain demonstrated in a long-term block plot trial series compared to initial assessments of genetic gain of the same families in small, young plots. We will further discuss the extension of genetic gain in multiple traits to biometric predictions of single-family stands at across the span of a rotation. Lastly, we will discuss the how the data from these studies provides an adjusted measure of genetic gain for deployed families compared to truly "wild" or "unimproved" stands.

WOODY DIVERSITY AND CARBON STOCK OF COCOA-BASED AGROFORESTRY SYSTEMS ON THE OUTSKIRTS OF FOREST CONCESSIONS IN SOUTHERN CAMEROON

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Cocoa cultivation, which is an important source of income for many households in Cameroon, is generally developed in a traditional way under the natural shade of the forest and combines various types of trees of socio-economic interest. The objective of this work is to assess the woody potential, uses and carbon stock of cocoa-based agroforestry systems on the outskirts of forest concessions in southern Cameroon. To do this, socio-economic surveys of 40 cocoa producers spread across 11 peripheral villages of UFA 09-026 and 09-027 were conducted for a sampling rate of 8%. These interviews were supplemented by floristic inventories carried out on 15 plots of 40 x 60 m for a total area of 15.84 ha. The data collected were subjected to various analyses and statistical tests (univariate test, analysis of variance, multivariate test). From our surveys 80% of respondents were men against 20% of women. The dominant types of estates are inherited farms (82.50%) and created farms (17.50%). The floristic potential is made up

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of 2776 individuals divided into 79 species, 69 genera, 31 families, and classified into 2 groups of woody plants, namely introduced and conserved species. Average density of cocoa trees is 1731.66 stems/ha against138.44stems/ha for associated species and 278 stems/ha in UFA forests. The Shannon index is 1.86 bits in SAFs versus 2.62 bits in UFA forests. Cocoa SAFs preserve 74.41% of conserved forest species and share 16.27% of these species evidence of the effectiveness of the conservation potential of these SAFs in the Bipindi area. *Persea americana, Elaeis guineensis* and *Mangifera indica* are the most ecologically important species while Malvaceae, Fabaceae and Apocynaceae are the most ecologically important families. *Ceiba pentandra* (506.31 tC/ha), *Terminalia superba* (106.21 tC/ha) and *Dialium pachyphyllum* (72.92 tC/ha) are the species that store the most carbon in the SAFs of Bipindi.

Session 2C: FOREST HEALTH

HISTORICAL GENETIC AND TREE STUDIES DIRECTORY

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Federal, state, and tribal governments spent millions of dollars during the mid-late 20th century to install and maintain thousands of genetic and tree improvement trials, includingseed orchards, throughout the eastern U.S. Many of these plantings were established to study the genetic and phenotypic variation for hardwood and conifer tree species. As time progressed, many of these plantings have been abandoned or forgotten due to the retirement of key scientists, lack of funding, a shift in priority, the degradation or loss of plantings, or the loss of data. Though some plantings have been removed, many still exist along with accompanying data. This untapped resource is in demand by 21st-century scientists to help overcome challenges like the lack of seed availability, and decimation of forests due to pests and to address key questions about assisted migration of forests. Locating these plantings and their data is essential to restore the services that these studies were established to provide. We were able to verify the existence of a list of historical plantings, by contacting affiliated scientists, using Google Earth Pro™, and visiting several universities and facilities to collect physical data. Across 48 states, 2,183 plantings have been confirmed. Of the 2,183, a total of 503 plantings were verified to be active; 268 were verified to not be recoverable; 48 were verified to be abandoned but not removed (inactive); and 355 have been confirmed to be active but lacking location information Out of the plantings that have been confirmed, only 629 have an accompanying dataset. Confirmed planting metadata are publicly available on an interactive dashboard. Current efforts include cataloging and tagging all files, even if they do not have an associated confirmed planting, to make the full dataset available to managers and researchers to play a part in retaining these datasets for future use.

A REGIONAL FOREST HEALTH COLLABORATIVE FOR TREE IMPROVEMENT BREEDING AGAINST INVASIVE PESTS AND DISEASES

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Eastern forests have been severely impacted by invasive insects and diseases culminating in decreased biodiversity, altered forest ecology, and reduced ecosystem services. Important riparian species such as the five native ash species (*Fraxinus* spp.) and American elm (*Ulmus americana*) are threatened with extinction because of an invasive pest (emerald ash borer) and disease (Dutch elm disease). Common northeastern forest types containing beech (*Fagus grandifolia*) and eastern hemlock (*Tsuga canadensis*) are under siege from beech bark disease, beech leaf disease, hemlock woolly adelgid (HWA), elongate hemlock scale. The Great Lakes Basin Forest Health Collaborative (GLB FHC), a partnership with Holden Forests & Gardens and the USDA Forest Service, was formed to help advance pest and disease resistance breeding for these important tree species. The primary mission is to establish a network of partners and provide training and technology transfer to them when needed. In turn, partners provide time and other resources to work together with the GLB FHC on activities including the identification of survivor trees with potential resistance, clone bank and/or seed orchard establishment, and seed collections; with the long-term goal of producing improved seed sources to restore impacted forests. We have partner networks for initial projects that include the identification and propagation of lingering ash and survivor elms, as well as seed collections for conservation. We anticipate expanding to projects in the future that involve breeding for HWA resistance and identifying beech trees that remain healthy in areas heavily impacted by beech leaf disease. Once networks are established for tree improvement programs, the GLB FHC partners can quickly mobilize to address new damaging pests as they arise.

ROLE OF SUPPRESSED RECOMBINATION IN THE ADAPTIVE RADIATION OF THE HAWAIIAN *METROSIDEROS*

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I will present how variation in recombination rate and inversion affects the genomic architecture of theHawaiian *Metrosideros* adaptive radiation. Recombination is essential for adaptation and speciation. It provides genetic diversity for populations and facilitates adaptation by bringing alleles into new combinations. However, breaking down co-adapted alleles associated with local adaptation would hinder adaptation and speciation when diverged populations connected via gen flow. Therefore, recombination suppression mechanisms would be favored to maintain linkage disequilibrium among co-adapted loci and act against homogenizing effects of gene flow. Inversions are well-studied mechanisms that suppress recombination leading to speciation. We thus predicted low recombining regions of the genome are enriched for inversions. The Hawaiian *Metrosideros* (Myrtaceae) is an example adaptive radiation of woody plant species. There are 25 taxa that are found non-randomly across the famously heterogeneous Hawaiian environment. The taxa have

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distinguishable morphological characteristics (leaf size and shape, pubescence) and reciprocal transplant experiments have shown each taxon are locally adapted to its endogenous environment. Population genomic studies have discovered genomic regions with elevated differentiation and interpret them as candidate regions for the adaptive radiation. We hypothesized genomic regions of elevated differentiation will have low recombination rate compared to background genome and they will be enriched for inversions. To test these two hypotheses, we constructed a genome-wide genetic crossover map for a F2 recombinant population generated from an outbred crossing scheme of two divergent *Metrosideros* taxa (glaberrima × incana) test and establish that suppressed recombination underlies the genomic basis of adaptive radiation in island organisms. We sequenced individuals using PacBio Hi-Fi reads representing the varieties included in the recombination map study, which was used to identify candidate inversions through comparative genomic analysis. Results evaluated the evolutionary role of recombination rate variation and inversions on the maintenance and selection of genetic variants during adaptive radiation.

QUERCHAOS: CHALLENGES OF CONSERVING A WEST TEXAS RED OAK SYNGAMEON

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Several West Texas relict oak taxa (*Quercus tardifolia, Q. graciliformis, Q. robusta*, etc) have been deemed of highest ex-situ conservation priority among all *Quercus spp.* in the United States. However, more intensive studies of these rare putative taxa are indicating some of these conservation targets may simply represent morphotypes within the hybrid gradient of a broad syngameon, within which other cryptic, reasonably stable undescribed species may be awaiting characterization. Being of great ecological importance to a region that has been naturally shifting to desertification since the Pleistocene, and further accelerated by human impacts, a syngameon-focused in-situ conservation approach is likely most effective to preserve adaptable genetics for the future than the taxon-specific limitations of ex-situ conservation endeavors.

Presentation Details

ADAPTIVE TRAITS OF WIDE-RANGING PROVENANCES OF LONGLEAF PINE (*PINUS PALUSTRIS MILL.)* GROWN IN THE NORTHERN EDGE OF ITS NATURAL RANGE: IMPLICATIONS FOR CONSERVATION, RESTORATION AND ASSISTED GENE FLOW

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Longleaf pine is native to the northern Gulf and southern Atlantic coastal plains of the United States. Due to heavy exploitation, it now covers only 3-5 % of its original land- base. In the state of Virginia, the northeastern leading edge of the species range, only 200 mature trees remain. A provenance test was established in Virginia using eight provenances, ranging from one “local” (VA) to sources extending to northern Florida and southern Mississippi. Unlike the other seven provenances, VA was derived from only 10 open-pollinated parent trees. VA emerged from the grass-stage fastest and exhibited higher survival through age 10 years. Mortality increased with the extreme minimum temperature (EMT) at the provenance origin. Of the surviving trees, there was no variation among provenances in growth rate. At age 10, VA had the highest yield index (m^3 ha^{-1}) with the relationship decreasing with provenance EMT. Cold tolerance was similarly related to provenance EMT. These results reflect adaptation to winter temperature along a clinal gradient. In stark contrast, VA had distinctly higher intrinsic water use efficiency than the other provenances and there was no relationship with provenance origin. Using DNA markers, VA had markedly lower allelic diversity compared to 15 other wide-ranging provenances tested including the ones used in our study and another from the state of Virginia. Although not inbred, VA showed curiously high degrees of relatedness (likely presence of full- and half-sibships) among the sampled trees and a strong skewness of parental contribution. Taken all together, the VA provenance and likely all extant native Virgina germplasm are invaluable for restoration and conservation efforts. Finally, as Virginia represents the leading northeastern edge of the species range, we consider if seed transfer guidelines for restoration purposes should be extended to include provenances that have evolved under warmer conditions.

Presentation Details

Day 4, Thursday, June 25, 2025

Session 3A: GENOMICS AND BIOTECHNOLOGY

GENOMIC CLUES TO PATAGONIA'S FATE: LOCAL ADAPTATION AND FUTURE MALADAPTATION IN *NOTHOFAGUS PUMILIO*

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The foundation tree species of the *Nothofagus* genus dominate South America's Patagonian forests, yet their patterns of local adaptation and climate change risk remain largely unexplored. The most widespread of these species, *Nothofagus pumilio*, has a range extending over more than 2,000 kilometers along the Andes Mountains, which feature two uncorrelated climatic gradients: a North-South temperature and day length gradient, and a West-East precipitation gradient. To identify signatures of selection along these gradients and characterize overall patterns of genetic diversity, we sampled 493 adults in 20 natural forest sites along the species' range in Argentina using a paired-site study design. Our landscape genomics analysis combined CHELSA climate data, 9,600 high-quality single nucleotide polymorphism (SNP) markers within fitness-related candidate genes, and several genome scan methods (LFMM, BayPass, PCAdapt, and pRDA). Signatures of selection were identified in hundreds of candidate genes, many of which are associated with temperature and day length factors and, to a lesser extent, precipitation. We then leveraged these results to predict future climate-driven maladaptation risk, by quantifying the necessary changes in allele frequencies to track changing climate conditions. Risk prediction results indicated a mosaic of maladaptation risk across the species' range, which suggests that the species' strong patterns of local adaptation could be detrimental in the future. Therefore, the persistence of *Nothofagus pumilio* within its current range is uncertain, which may have dire consequences for Patagonian forests as a whole.

THE ROLE OF TELOMERES IN LONG-LIVED TREES: ARE THEY A BIOMARKER FOR ENVIRONMENTAL STRESS?

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Telomeres are repetitive G-rich sequences of DNA found at the ends of chromosomes that act to protect chromosomes from DNA damage. Telomeres play an important role in maintaining genome stability and their loss has been associated with aging and physiological stress in mammalian systems. Telomere attrition can occur during cellular replication; however, this attrition can be counteracted by activity of the telomerase enzyme which acts to maintain telomere length (TL) homeostasis. In plants, natural variation in TL has been linked to the evolution of life history strategies critical to adaptation, suggesting disruption of TL homeostasis can have detrimental effects on an individuals' persistence or fitness. However, factors influencing TL variation and its relationship to fitness in plants remain largely untested. In this study, we evaluate TL variation as a predictor for responses to environmental stress and examine the relationship between TL loss and life history strategies in a long-lived model system. In addition, we quantify the contributions of genetics, environment, and genotype-by environment interactions to TL variation. *Populus* provides a model forest tree system to study TL dynamics due its rapid growth, ability for clonal propagation, and ecological and economic importance. Leveraging replicated common garden experiments of *P. trichocarpa*, *P. balsamifera*, and their hybrids planted across North America, we explore the relationship between TL change and climate transfer distance. Genotypes were sampled at the beginning and end of the growing season, and qPCR was used to measure the rate of change in TL. We used climate transfer distance to predict the rate of TL change through the growing season. In addition, we quantified the relationship between TL and life history traits important to adaptation. These data will provide new insights into the role of telomeres as a potential biomarker to indicate plant response to environmental stress.

GENE EXPRESSION IN EASTERN OAK SPECIES IS IMPACTED BY SOIL NUTRIENT PROFILE

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Climate adaptation has emerged as a priority in forest management as projected changes in climate are expected to impact forest ecosystems. In Pennsylvania, some species will thrive under changing conditions, others may decline, and new species may establish. However, these projections do not account for interactions with belowground environments, including nutrient availability, pH, and water availability which may also impact the success of species. Previous studies have suggested that some trees accumulate more biomass on sites with higher or lower nutrient availability depending on their survival and growth strategies. This experiment explores the interactions of different belowground environments with native and introduced tree species. One thousand seedlings from a mixture of five native species and four novel species were planted across four different common garden sites with a gradient of soil nutrient profiles. We measured seedling health and growth twice a year for four growing seasons (spring 2021-fall 2024). In August 2021, leaf material was collected from three native oak species, northern red oak (*Q. rubra*), black oak (*Q. velutina*) and white oak (*Q. alba*), and one novel, future climate adapted species (southern red oak - *Q. falcata*), at two different plots with significantly different availabilities of key macronutrients to evaluate gene expression differences potentially attributed to species-

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specific interactions across varying belowground environments. A weighed Gene Correlation Network Analysis was performed to evaluate patterns of co-expression among the most differentially expressed genes. Across the two plots, genes associated with regulation of cellular processes and stress responses, as well as mechanisms for regulation of ion-binding and photosynthesis were differentially expressed. Quantifying variation in gene expression across different species under varying soil nutrient profiles will aid in identifying the genetic mechanisms underlying adaptation to belowground environments. Ultimately, this work will be critical to development of assisted seed transfer guidelines under climate change.

A GENOMIC PORTRAIT OF REGIONAL INTROGRESSION IN *QUERCUS MACROCARPA*

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Adaptive introgression, the movement of beneficial genes from one species into another species through hybridization and backcrossing, can provide genomic diversity which can be harnessed to identify resilience to both biotic and abiotic factors. The eastern North American white oak syngameon is composed of over a dozen *Quercus* species many of which co-occur and can interbreed, resulting in viable offspring. This syngameon includes the widespread and morphologically variable *Q. macrocarpa*. We used range-wide genome resequencing data from *Q. macrocarpa* and co-occurring interfertile white oak species to explore the geographical and genomic landscape of introgression and hybridization. We found that hybridization is not uniform across the range of *Q. macrocarpa*, but its frequency varies geographically. On the other hand, we found that introgression has occurred across the geographical range and across the genomic landscape. However, introgression disproportionately affects some areas of the genome over others.

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A PAN-GENOMIC VIEW OF INTROGRESSION IN HYBRID POPLARS

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Presence-absence variation (PAV) underlies phenotypic variation within and between many species, but the evolutionary forces that shape standing PAV remain poorly-understood. In forest trees, interspecific hybridization is common, and introgression may be a source of PAVs that impinge or promote the evolution of adaptive traits. Here, we used PacBio long-read sequencing of 40 individuals to detect and characterize PAV segregating within the natural hybrid zone between *Populus balmsamifera* and its recently diverged sister taxon, *Populus trichocarpa*, which occupy distinct climatic niches. These two species naturally hybridize where their ranges overlap, and introgression may contribute to local adaptation along climatic and other environmental gradients. Nearly all population genomic studiesof *P. trichocarpa* and *P. balsamifera* have mapped reads back to a single reference genome (Nisqually-1) to identify segregating genomic variation. Thus, PAV not present in Nisqually-1 (including those unique to *P. balsamifera* or hybrid zones) may have been overlooked by prior studies of climate adaptation for these species. We constructed a pan-genome graph from alignments of de-novo genome assemblies for 16 samples of *P. balsamifera*, *P. trichocarpa* and admixed genotypes sampled from hybrid zones in Alaska and the Pacific Northwest. We aligned short-read sequences from over 500 additional individuals to this pan-genome graph in order to genotype PAV segregating within natural *Populus* hybrid zones. Using a combination of selections scans, local ancestry inference and genotype-phenotype associations, we identified PAV that may contribute to the genetic and ecological divergence between *P. trichocarpa and P. balsamifera*, as well as variants that may play a role in adaptive introgression between these species. We also explore the potential functional consequences of PAV segregating within natural hybrid zones. Our results shed light on the porosity of species boundaries with respect to structural variation and clarify the role that adaptive introgression plays in shaping the distribution of PAV across admixed populations.

GENETICS- ENABLED RESILIENT LODGEPOLE PINE TREE IMPROVEMENT STRATEGIES

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The productivity and sustainability of forest ecosystems and operations depend on healthy and resilient forests. Warming since the 1850's has compromised this resilience, exacerbating biotic and abiotic threats, often in combination. Our studies investigated the impact of warming on resistance phenotypes in both univariate and multivariate adaptive spaces using tree heights as a proxy for fitness. Across 15 traits studied, cross-site heritability was higher for growth characteristics, such as height and DBH, compared to wood quality and resistance attributes, including as wood density, dendrochronological growth decline index (DECL), stable isotope ratio ($\delta^{13}C$) and defense compounds. Overall, 204 genomic variants are found associated with phenotypic variability across all sites with a multi-trait association mapping approach, while conventional uni-variety methodology detected only 27 associated variants. Although dominance variance accounted for less than 14% of the total genetic variance on average, non-linear kernel predictive algorithms, capable of capturing complex genetic architectures, outperformed linear predictors in terms of both accuracy and bias. Our results suggest seed source populations from warmer climates possesses greater adaptive capacity than those from cooler regions. Based on our results, in a warming scenario, directional selection could increase adaptability for drought avoidance phenotypes ($\delta^{13}C$), whereas host tree's suitability to the mountain pine beetle was subject to divergent selection, with the lowest response to selection in a warmer, higher rainfall climate. By comparing seed-origin populations of lodgepole pine across test sites that simulate future *in situ* climate adaptation scenarios, we highlight the potential effects of climate change on forest resilience and adaptation strategies.

Session 3B: SYMPOSIUM ON NORTH AMERICAN ASH

PROGRESS IN EAB-RESISTANCE BREEDING PROGRAMS

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The invasion of emerald ash borer (EAB, *Agrilus planipennis*) threatens the survival of ash species (*Fraxinus)* in the United States. Imperiled species include *F. americana*, valued for hardwood timber, and ecologically important species, particularly in riparian and wetland forests (e.g., *F. pennsylanica* and *F. nigra*). We define “lingering ash” trees as mature ash trees in natural forests invaded by EAB that maintain healthy canopies for at least two years after all others have died. EAB egg bioassay experiments on grafted ramets of lingering green (*F. pennsylvanica)* and white (*F. americana)* ash selections confirmed that up to half of these trees possess an increased level of resistance due to several types of defense responses, including mortality of early instar larvae. Bioassays also demonstrated that many seedling progeny from controlled cross-pollinations

between lingering ash genotypes have as much or more EAB-resistance than their parents. Furthermore, estimates of genetic heritability predict sufficient genetic gain through breeding. Field trials have been established to validate tree performance with the longer-term goal of converting plantings into seed orchards by removing poor performing trees. A pilot-study screening unselected black ash (*F. nigra*) seedlings for EAB resistance suggests that a pro-active breeding approach has promise, but bioassay methods and lingering black ash selection criteria will require additional optimization. Bioassays on additional black ash seedlings from a wider geographic area are planned to confirm our initial findings.

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FIELD PLANTINGS TO ASSESS EAB RESISTANCE IN THE USFS ASH BREEDING PROGRAM

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In addition to the propagation and greenhouse testing facilities of the US Forest Service Ash Breeding Program in Delaware, Ohio there are several plantings located at the Delaware site, and at partner sites in Ohio and adjacent states. Planting types include both archive and experimental with several plantings designated to become seed orchards. Early program goals included understanding ash resistance to EAB in non-native species, especially co-evolved Asian species. A total of five plantings have been established at three sites in Ohio to assess resistance across these species. Some Asian species have excellent EAB resistance and appear to be thriving in the mid-Ohio region, while others were unable to survive winter cold and EAB infestation. Beginning in 2008, the ash breeding program shifted focus to "lingering" ash, trees that had survived peak mortality caused by EAB. Two replicate lingering ash clonal trials have been established to confirm field resistance and correlate egg transfer bioassay to field performance of these selected trees. Establishing heritability of resistance in the lingering ash is essential, so controlled pollinations using lingering ash selections as parents were conducted. The resulting progeny families have been tested in both egg transfer bioassays and/or planted in progeny test plantings, including a unique approach to allow 'stumps' of seedlings that underwent destructive bioassays to resprout and be planted for field assessment. Unscreened siblings have been planted at additional sites, and all plantings are accumulating EAB damage from natural infestation. Additional plantings have been established to assess baseline resistance in wild black (brown) and blue ash and demonstrate reintroduction of ash in park settings. Challenges to data analyses include differences in phenotypes between species (including different physiology of resistance), choosing the best metric to efficiently rank parents, and integrating field planting-based results with egg transfer bioassay results and rankings.

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GENOME SIZE AND PLOIDY VARIATION IN *FRAXINUS*

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Conservation efforts in North American *Fraxinus* are hampered by problems with species definitions. Flow cytometry surveys have been carried out on *Fraxinus* spp., with emphasis on the North American species. Across the genus, haploid genome size (1C value) varies from 0.7-1.0 pg, but shows little variation within the six sections. Polyploidy is frequent only in the North American sect. *Melioides*, where ploidy levels run from diploid to octaploid. Outside sect. *Melioides*, polyploids are found only in some populations of the unrelated *F. chinensis*. The two clades that contain polyploids are both dioecious. Surveys using a combination of ploidy measurements and molecular phylogenies are leading to well-supported species definitions in sect. *Melioides* that reflect population structure and genetic relatedness. In particular, it is clear that *F. biltmoreana*, *F. pauciflora*, and an undescribed tetraploid species are distinct from *F. americana*, and conservation work in the central and southeastern US must separate these species.

THE EVOLUTION OF SAMARA MORPHOLOGY DIFFERENCES IN *FRAXINUS NIGRA*

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Life history variation at the juvenile stage can have long-term impacts on individual reproduction and survival for forest tree species. Selection at the seed and seedling stage dictates the environmental space an individual occupies for its entire lifetime. Therefore, understanding how selection has shaped evolution of genetic differences in seed and seedling traits that may impact fitness is crucial to predicting how populations will establish and persist under climate change. We examined differences in samara morphology for seeds sourced from across the distribution of black ash (*Fraxinus nigra*) to estimate how variation in morphological traits is partitioned between and within populations. We leveraged existing black ash collections from 694 maternal families representing 75 provenances. A subset of 50 samaras per family were x-rayed, and seed and wing components were segmented using a patch-wise, attention gated, 3D fully convolutional machine learning algorithm. Using linear mixed effect models, we estimated the proportion of trait variation explained by differences between and within populations and quantified potential trade-offs associated with resource allocation and dispersal between seed and wing tissue. To test the effect of variation in historic maternal climate on variation in samara morphology, we used partial redundancy analysis. A reasonable proportion of morphological variation for black ash samaras is explained by provenance (~10%) while a substantial proportion is explained by within population differences (36-55%). Redundancy analysis indicates that on average, while controlling for geographic proximity, morphological trait differences can be explained by maternal climate. The variation in

The variation in maternal climate has likely contributed to differences in samara morphology at the population level, with larger seed and wing traits associated with historically warmer climates. However, substantial within population variation suggests there is diversity for selection to act upon at this critical life stage to ensure the evolutionary potential of the species long term.

POLYPLOIDY AND HYBRIDIZATION AMONG *FRAXINUS* SPECIES IN THE GREAT SMOKY MOUNTAINS NATIONAL PARK

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The sudden endangerment of North American ash (*Fraxinus* spp.) by emerald ash borer (EAB) has prompted extensive efforts to characterize and breed resistance among rare, partially resistant individuals. However, substantial knowledge gaps remain in the fundamental ecology and biodiversity of ashes across the New World. Recent genomic evidence suggests that polyploidy and cryptic hybridization remain significant obstacles to effectively conserving the dwindling, wild genetic diversity within *Fraxinus*. Unfortunately, little time remains to characterize these dynamics due to the widespread mortality caused by EAB. In this study, we assess how dioecious species of ash in section *Melioides* hybridize in the Great Smoky Mountains National Park, a UNESCO world heritage site and temperate biodiversity hotspot. We explore how local, microclimatic barriers may impact gene flow among three species of ash [white ash (*Fraxinus americana*; 2n), green ash (*Fraxinus pennsylvanica*; 2n), and Biltmore ash (*Fraxinus biltmoreana*; 6n)] across fine spatial scales through ecological niche modeling. We report compelling evidence that gene flow among ash species does not only occur among diploid congeners, but also across ploidies, based on whole-genome resequencing data (n = 367 trees). Furthermore, this study provides evidence that Biltmore ash is substantially more ubiquitous in the Blue Ridge Mountains ecoregion than previously recorded, suggesting that ash biodiversity may be underestimated range wide. By combining forest health surveys, genomics, and ecological modeling, we describe niche variation and survivorship among white, green, and Biltmore ash, respectively, in a highly heterogeneous mountain ecosystem at 30-m spatial resolution. These results will inform both local conservation management and add to the growing evidence that ash species diversity and hybridization is underestimated across North America.

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MUCH ASSEMBLY REQUIRED: AN OCTOPLOID GENOME OF CONSERVATION CONCERN

Christopher Guzman [1], David Baukus [1], Laurel Humphrey [1], Owen McEwing [1], Emily Trybulec [1], Karl Fetter [1], Michelle Neitzey [1], Cynthia Webster [1], Nicole Pauloski [1], Zane Smith [2], Meg Staton [2], Rachel O'Neill [1], and Jill L Wegrzyn [1]

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All North American ash populations have been severely impacted by the invasive, wood-boring beetle, known as the emerald ash borer (*Agrilus planipennis*, EAB). *Fraxinus profunda* (pumpkin ash), like most North American ash species, is classified as critically endangered by the International Union for Conservation of Nature (IUCN). The narrow geographical range and low genetic diversity of this species make it particularly vulnerable to population decline. Genome assembly is a crucial first step in developing a reference that can improve population diversity estimates, examine the species' evolutionary history, and identify genomic regions that may contribute to EAB resistance. However, a major challenge in this assembly is the complex and mysterious origins of its ploidy. Recent estimates suggest it is an octoploid, initially believed to be a hybrid product of tetraploid *F. americana* and diploid *F. pennsylvanica*. However, recent range wide genotyping suggests that *F. caroliniana* and *F. pauciflora* may be more closely related. To generate the first reference genome for *Fraxinus profunda*, we assembled Oxford Nanopore and PacBio HiFi long reads and scaffolded the assembly using Illumina sequenced Hi-C. Here, we discuss the strategies involved in assembling complex and recent polyploid genomes, as well as the significance of this reference in furthering conservation genomic research.

INTEGRATING TRADITIONAL BREEDING AND BIOTECHNOLOGY FOR RESILIENT URBAN ASH TREES

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Urban environments present multiple tree stressors, including soil compaction, limited water availability, and pollution. The Daniel P. Haerther Charitable Trust New Plant Development Program (NPDP) at Morton Arboretum addresses these challenges by breeding and researching woody landscape plants adapted to urban conditions. Established in 1968, our program builds on Dr. George Ware's ecological niche approach. It investigates plants from natural settings that demonstrate traits valuable for human-modified landscapes. Our current research focuses on *Fraxinus* species resistance to the emerald ash borer (EAB), employing an integrated approach through collaboration with various bioassays, gene analysis, and traditional breeding. We have established in vitro clonal germplasm to identify genetic

characteristics associated with resistance. Our work in embryogenic culture systems promises to change how we produce and study resilient ash trees. These advanced bioreactor-based systems will rapidly scale genetically superior germplasm, potentially reducing development timelines from decades to years and providing a common basis for experimenters across disciplines to gain access to a common standard set of interesting clonal cultivars. We are developing micropropagation techniques and applying functional genomics to identify regulatory genes critical for EAB resistance. Our molecular approaches include differential gene expression analysis using ddPCR for genes of interest, and we are establishing baseline gene expression profiles for *F. mandshurica*, *F. pennsylvanica*, and *F. americana*. Additionally, we are developing *Agrobacterium* transformation protocols to support biotechnology applications for understanding resistance mechanisms. This integrated approach aims to accelerate breeding cycles, enhance conservation efforts, and improve the long-term survival of *Fraxinus* species in urban environments facing increasing biotic and abiotic stressors.

GENOMICS-DRIVEN MONITORING OF *FRAXINUS LATIFOLIA* (OREGON ASH) TO INFORM CONSERVATION AND EAB-RESISTANCE BREEDING

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Understanding the distribution of standing genetic variation for species of conservation concern is critical for designing conservation and restoration strategies. Genetic data can provide baseline knowledge needed to establish or prioritize conservation, restoration, or pre-breeding strategies. In this study, reduced representation sequencing was used to generate genomic data for over 1,000 samples from 61 populations across the range of *Fraxinus latifolia*, a foundational riparian tree native to the Pacific Northwest recently exposed to the invasive Emerald Ash Borer (*Agrilus planipennis*), to inform the establishment of genecological resources needed for proactive conservation and breeding. Populations were genetically structured along a latitudinal gradient, with population connectivity largely maintained along central valley river systems and a center of genomic diversity within the Columbia River watershed of Washington and Oregon. Interestingly, putative polyploids were identified in southern California that may represent tetraploid hybrids. Despite evidence of gene flow, estimates of nucleotide diversity and effective population size estimates were low across all populations,

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suggesting that the patchy distribution of *F. latifolia* across parts of its range may impact long-term evolutionary potential of the species. In addition, range-wide genetic offset suggests populations may be at further genetic risk under climate change. Targeted *ex situ* collection strategies to preserve both core and peripheral populations across the range will capture a majority of genetic diversity, and propagation of germplasm from four recommended conservation units will preserve variation needed to initiate range wide EAB-resistance breeding trials for *Fraxinus latifolia.*

MULTI-YEAR ANALYSIS OF THE CHEMICAL DEFENSE DEPLOYED BY GREEN ASH (*FRAXINUS PENNSYLVANICA)* AGAINST EMERALD ASH BORER *(AGRILLUS PLANIPENNIS)* AND THE CHANGES IN GENE EXPRESSION ASSOCIATED WITH SUCCESSFUL DEFENSE

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The emerald ash borer (*Agrilus planipennis*, EAB) invasion in North America threatens most North American *Fraxinus* species, including green ash (*F. pennsylvanica*), the mostly widely distributed species. A small number of green ash (0.1-1%) survive for years after all surrounding green ash have been killed by EAB and kill more EAB larvae when challenged in greenhouse studies. We combined untargeted metabolomics and associative transcriptomics with intensive phenotyping in three sets of full-sib progeny having susceptible or lingering ash parents to detect chemotypes and differentially expressed genes associated with defensive responses to EAB. The phenotypic distribution (0-100% of larval killed) of progeny from lingering × lingering full-sib families and a susceptible full-sib control, combined with untargeted metabolomics profiles from these same full-sib families, provided insights into the effective and ineffective responses deployed by green ash against EAB. We detected select secoiridoidss associated with an ineffective response to EAB infestation (low larval kill) and alkaloids that may have a role in an effective defense response. We have also identified differentially expressed features in the transcriptome associated with a response to infestation and high % larval kill, providing future avenues to explore the effective defense deployed by select green ash against emerald ash borer. Our data indicates that EAB infestation induces a defensive response in every tree but only a few trees mount an effective defense, killing enough early instar EAB larvae to prevent lethal damage to the vascular system.

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Session 3C: SYMPOSIUM ON MOLECULAR APPLICATIONS IN TREE BREEDING

GENOMIC RESOURCES FOR LOBLOLLY PINE GENE DISCOVERY AND BREEDING

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Our objective is to assemble and annotate a high quality, chromosome-level reference genome for loblolly pine. Using ultra-long nanopore reads at 25x coverage, we assembled a draft genome with 59,168 contigs totaling 24.7 Gb with an N50 contig size of 1.83 Mb increased from 25.36 Kb in v2.0 and the N50 scaffold size increased from 107.04 Kb to 55.1 Mb with a BUSCO of 86%. We also generated ~100x coverage with PacBio HiFi of the same genotype, providing high base-pair accuracy for phased assemblies. A preliminary assembly of with the HiFi data resulted in 4312 total contigs (1,875 over 50 Kb) totaling 20.4 Gb. Our progress integrating the long read nanopore, HiFi and Omni-C sequence data to improve scaffolding and ordering of contigs to develop a high-quality chromosome-level assembly will be presented. We also integrated all available data to create a new loblolly pine transcriptome of ~50,000 transcripts to facilitate gene calling and annotation, especially in complex regions with repetitive sequences and structural variations. With the new loblolly pine transcriptome, loblolly v2.01 genome and a new de novo slash pine transcriptome we identified a large set of NLR gene families, some of which are predicted to be important in defense against pathogens and insect pests. We categorized the domain structure and designed probes for R-gene capture sequencing. We plan to present the results of R-gene capture sequencing with a wide selection of loblolly pine trees from across the natural range to test our hypothesis that individuals in the western population contain a greater number of NLR genes than individuals in the eastern population.

VALIDATION OF GENOMIC SELECTION ACROSS THREE GENERATIONS IN *Pinus taeda*

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Presentation Details

The North Carolina State University Cooperative Tree Improvement Program has developed a multi-generational closed breeding population with small number of founders (n=21) to validate genomic selection (GS) in *Pinus taeda*. The first generation was comprised of 2400 cloned individuals originating from 51 full-sib and 7 half-sib families that were field tested at eight locations in the southern US. The second generation was composed of 73 first generation clones that were mated to produce 67 full-sib crosses. The seedling progeny of the second generation were planted at four locations in the southern US. The first- and second-generation trials were assessed at age six and four respectively for growth, disease resistance and stem quality. About 2100 trees from each generation and their pedigree were genotyped using the Pita50K Axiom SNP array developed for *P. taeda*. The first-generation cloned population (n=2,100) was used for model training. The seedling progeny of the second-generation population (n=2,100) was used for model validation. A single-step GBLUP model was applied to predict genomic estimated breeding values (GEBVs) for height, volume, and stem straightness in the validation population. GEBVs were compared with breeding values estimated using conventional pedigree BLUP (EBV). The predictive ability of SNP markers to predict GEBVs was 0.47 for height, 0.52 for volume, and 0.58 for stem straightness. The prediction abilities for the above complex traits using SNP markers were similar to the confidence of conventional selection (squared root of heritability of the traits). Without ambiguity, the results validated the efficiency of GS in a conifer forest tree. Plans are underway to implement GS in *P. taeda* starting in 2026.

PEDIGREE ASSIGNMENT USING LIKELIHOOD GENOMIC RELATIONSHIPS IN A BREEDING POPULATION OF *PINUS TAEDA*

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Conventional breeding programs have historically relied on pedigree records to estimate genetic relationships and manage diversity within breeding populations. Pedigree information also guides crossing decisions to optimize genetic gain while controlling inbreeding. However, pedigree errors are common in plant and animal breeding programs and, if uncorrected, can lead to genetic losses and increased inbreeding. DNA markers enable accurate parentage assignment and improve genetic relationship estimates for breeding applications. In this study, we used the Pita50K SNP array to genotype two generations of a *Pinus taeda* breeding population and assess pedigree accuracy. Parentage assignment was performed using the Genomic Relationship Likelihood (GRL) software, which applies to a likelihood-based approach to genomic relationships derived from SNP markers. The pedigree error rate was 1.7% due to mislabeling 34 progeny out of 2000. To enhance accessibility, we are currently developing an R package to facilitate the use of this software in a user-friendly manner.

Presentation Details

ADVANCING MARKER-ASSISTED SELECTION FOR BLIGHT RESISTANCE IN HYBRID CHESTNUT

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The restoration of the American chestnut (*Castanea dentata*) has been challenged by the complex genetic basis of blight resistance. In this study, we refined the genetic model of resistance using enhanced genome-wide association study (GWAS) methodologies, specifically FarmCPU and BLINK algorithms. These approaches identified twenty peak quantitative trait nucleotides (QTNs). Eleven QTNs were associated with well-defined quantitative trait loci (QTLs), collectively explaining up to 23.2% of the variance in canker size while occupying only 5% of the genome. Many of these QTLs are enriched for nucleotide-binding leucine-rich repeat (NLR) genes, as well as genes involved in reactive oxygen species (ROS) regulation and systemic acquired resistance (SAR) pathways, offering new insights into resistance mechanisms. To validate our findings, we analyzed a large dataset of genotyped and phenotyped hybrid chestnut trees curated by The American Chestnut Foundation (TACF). Our results indicate that marker-assisted selection (MAS) could facilitate the stacking of resistance QTLs with alleles derived from Chinese chestnut (*Castanea mollissima*), while conserving American chestnut alleles elsewhere in the genome. This strategy may enable the development of backcrossed individuals with both high blight resistance and predominantly American chestnut ancestry, advancing restoration efforts.

Session 4A: GENOMICS and BIOTECHNOLOGY

GENETIC AND ENVIRONMENTAL DRIVERS OF INTRASPECIFIC VARIATION IN TREE LEAF PHENOLOGY

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Presentation Details

To understand how trees will respond to changing climates, it is crucial to characterize the genetic population structure of tree species and how these genetic patterns shape phenotypic variation in relation to climate. One phenotype of particular importance for reforestation and forest regeneration efforts is the timing of life-cycle events, or phenology. Shifts in climate over recent decades have advanced the onset of spring and delayed fall phenology, resulting in longer growing seasons. While day-length and temperature cues primarily govern current phenological variation, we lack understanding of how plants will evolve as environmental factors influencing traits like spring leaf-out and fall senescence increasingly uncouple from day-length cues. Understanding how genetic variation underpins phenological traits, and how these traits vary across a species' range, is vital for predicting plant responses to climate change. I share data I collected from 550 individual trees across four species at Black Rock Forest over four seasonal transitions from Fall 2023- Spring 2025, noting the effects of an extremely wet and then dry year. This enables the parsing out of microenvironmental factors influence in phenological traits like leaf-out timing. I also focused on sugar maple (Acer saccharum), an ecologically and economically significant species, to investigate the genetic underpinnings of phenological variation in this species. Genomic information for a subset of individuals from the Black Rock Forest population, along with genomic data from 10 other sugar maple populations, was used to map population genetic structure and assess how genetic diversity influences phenotypic variation in response to climate.

ASSISTED MIGRATION NEEDS OF NORTHERN RED OAK (*QUERCUS RUBRA*)

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Intensifying climate change exacerbates droughts, heatwaves, and extreme weather events, endangering global forest health and biodiversity. Assisted migration is increasingly recognized as a management strategy for safeguarding forest species. But simply moving tree populations towards expected future mean annual temperatures does not account for increased extreme weather events nor incorporate trees' phenotypic plasticity. In 2019, we planted a replicated provenance trial with 19 populations and 118 open-pollinated families of northern red oak (*Quercus rubra*) spanning 10°C mean annual temperature (MAT 4.1°C - 14.2°C). The provenance trial sites are located near the northern seed collection limit (Ford Center and Forest, MAT 4.4°C) and at a central location (W.K. Kellogg Experimental Forest, MAT 8.8°C). To assess the performance of populations within and beyond their natural distribution, we measured annual growth, tree and leaf morphology, leaf and root physiology, foliar metabolites, pest and pathogen damage, plant phenology, and frost damage. We assessed phenotypic plasticity by comparing measured traits at both sites within families, and traits measured on the same trees over time. Variance components and trait heritability were estimated. Traits related to physiology showed some phenotypic

plasticity, allowing trees to adapt to varying environments. Variance component analysis indicated high genetic variability within populations except for a few traits such as fall phenology and growth in years with extreme weather events. While extreme weather events had a minimal effect on populations shifted a few degrees Celsius north, the phenology of trees from much warmer seed sources was not adapted to longer northern winters, leading them to suffer during unseasonal frost events. Northern seed sources exhibited conservative growth strategies, while southern populations adopted acquisitive strategies that often led to bush-like growth after multiple rounds of frost damage to the leading shoot.

PROVIDING BIOLOGICAL CONTEXT FOR GWAS RESULTS USING EQTL REGULATORY AND CO-EXPRESSION NETWORKS IN *POPULUS*

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Our study utilized genome-wide association studies (GWAS) to link nucleotide variants to traits in *Populus trichocarpa*, a species with rapid linkage disequilibrium decay. The aim was to overcome the challenge of interpreting statistical associations at individual loci without sufficient biological context, which often leads to reliance solely on gene annotations from unrelated model organisms. We employed an integrative approach that included GWAS targeting multiple traits using three individual techniques for lignocellulose phenotyping, expression quantitative trait loci (eQTL) analysis to construct transcriptional regulatory networks around each candidate loci, and co-expression analysis to provide biological context for these networks, using lignocellulose biosynthesis in *Populus trichocarpa* as a case study. The research identified three candidate genes potentially involved in lignocellulose formation, including one previously recognized gene (Potri.005G116800/VND1, a critical regulator of secondary cell wall formation) and two genes (Potri.012G130000/AtSAP9 and Potri.004G202900/BIC1) with newly identified putative roles in lignocellulose biosynthesis. Our integrative approach offers a framework for providing biological context to loci associated with trait variation, facilitating the discovery of new genes and regulatory networks.

Presentation Details

Presentation Details

A PRELIMINARY ASSESSMENT OF THE FUTURE ADAPTABILITY OF EASTERN BLACK WALNUT (*JUGLANS NIGRA* L.) USING POPULATION GENOMIC VARIATION

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Numerous studies in forest trees have shown that genetic variation is arrayed along the environment as a result of post-glacial phylogeography. These locally adaptive patterns of variation were driven by demography and selection over vast stretches of time. In contrast, the disturbances attributed to climate warming threaten to disrupt these adaptations abruptly and unevenly across the landscape. For scientists, land managers and other stakeholders, a foremost goal is to mitigate the effects of these disruptions through assisted migration, climate-smart seed sourcing and identifying innate adaptations to novel environments. An initial step in this endeavor is to accurately predict the relative degree to which parts of species' distribution may experience disruption of local adaptation. A novel metric, *genomic offset,* employs data on contemporary population genomic variation and current and future climates to reliably predict zones of population These genomic predictions are then validated using growth and fitness data from common garden studies and are beginning to be used in various mitigation strategies that broadly fall under the umbrella of assisted migration. Eastern Black Walnut is highly prized for timber and there is immense interest in its commercial cultivation. Despite this, information available on locally adaptive genetic variation in natural populations is limited. We studied genomic variation in Single Nucleotide Polymorphisms (SNP) in 95 individuals from 12 populations of *Juglans nigra* distributed along four regional quadrants (ne, nw, se, sw) encompassing nine states. We associated these SNPs with bioclimatic variables (WorldClim) to identify variants likely under local adaptation to the environment. Then we used the machine learning algorithm *Gradient Forest*to estimate *genomic offsets* in order to predict where, and to what extent these *GxE* relationships were prone to disruption in 2070 and beyond. Results are discussed within the context of restoration and development of climate-smart seed sourcing strategies.

RIBOSOMAL GENE EVOLUTION IN CHESTNUT: A CYTOGENETIC PERSPECTIVE ON ADAPTATION

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Evolution continuously shapes species over time. Fossil records suggest that chestnuts (*Castanea*) and oaks (*Quercus*) diverged from a common Fagus-like ancestor around 60 million years ago. The American chestnut (*C. dentata*) separated from the Chinese chestnut (*C. mollissima*) approximately 3–5 million years ago, likely due to climatic and geographic factors. Once known as the "King of the Appalachian Forest," the American chestnut was nearly wiped out by chestnut blight, caused by the fungal pathogen *Cryphonectria parasitica*. The American Chestnut Foundation (TACF) has been working to transfer blight resistance genes from Chinese chestnut. The U.S. Forest Service's SRS-4160 Cytogenetics Unit teamed up with TACF to provide critical cytogenetic insights to accelerate the development of blight-resistant American chestnut. Our fluorescence in situ hybridization (FISH) analysis confirms that both species have two 35S rDNA loci (one major, one minor) and a single 5S rDNA locus, each located on distinct chromosomes. Recently, we identified a Chinese chestnut accession where the minor 35S rDNA locus exhibits an unusually high copy number, resembling a second major site. This suggests an ongoing evolutionary process, potentially driven by environmental pressures, in which a minor locus may transition into a major one. By identifying homozygous individuals through FISH and comparing them to normal variants, we can explore the functional implications of this shift. While such changes typically take millions of years, our research provides real-time insights into the evolutionary dynamics of plant genomes.

SESSION 4B: DIGITAL TOOLS, NETWORKS AND MODELS

A DEEP LEARNING APPROACH TO SPECIES DISTRIBUTION MODELLING FOR NORTH AMERICAN TREE SPECIES

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Presentation Details

Selecting suitable tree species and seed sources for future climates requires reliable models of tree species distributions. We developed a deep neural network approach to estimate the range and abundance of major North American tree species, supporting the North America Seed Selection Tool (http://tinyurl.com/NA-SST). The web tool is meant guide climate-informed forest management and facilitate cross-border seed transfers. We combined forest inventory and ecological plot data from Canada, the United States, and Mexico for all major North American tree species. Misidentifications and out-of-range records were removed by cross-referencing Little's historical distribution maps with a 200km buffer for spatial inaccuracies. Predictor variables include historical climate normal data (1951–1980) for 12 bioclimatic variables at 1km resolution as well as topographic indices (e.g., slope position, exposure, etc) as well as remotely sensed land cover. The neural network models interactions among climate, terrain, and land cover classes, producing a frequency estimate reflecting the relative basal area of species, with two outputs: (1) species range maps in a sense of climatically suitable habitat conditions, and (2) actual species distribution maps that are filtered by remotely sensed deciduous and coniferous tree cover. The model output that delineates the climate habitat are integrated in the NA-SST web application, allowing forest managers to select candidate species and locate suitable seed sources under observed and projected climate change.

ENHANCING FOREST RESEARCH WITH TREESNAP AND TREESNAP PLOTS: MOBILE PLATFORMS FOR DATA COLLECTION AND PHENOTYPING

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Forest research often relies on both professional researchers and citizen scientists to gather observational data and physical samples. To improve this process, we have developed two mobile and web apps: **TreeSnap**, to tag individual trees, and **TreeSnap Plots**, for plot monitoring. These platforms guide users in collecting uniform, high quality data and samples, increasing both data accuracy and research efficiency. TreeSnap launched in 2017 and helps the public identify trees of interest to partner scientists. Often, these are trees with notable resistance to endemic pests and diseases, such as healthy ash, chestnut, hemlock, and elm. TreeSnap includes species-specific questionnaires and images to guide phenotyping, enhancing data utility for researchers and ensuring valid data from volunteer citizen scientists. With over 3,000 active users and 22,000+ trees tagged, TreeSnap has been utilized by 18 partner research organizations or projects. The app works globally, even in places without cellular service, storing data locally until network or wifi connectivity is available for syncing. After data upload, TreeSnap's web interface offers

a public data dashboard and a private researcher portal for filtering, searching, and downloading datasets. We have also developed **TreeSnap Plots** to support long-term forest health monitoring. This app will allow researchers to set up flexible plot monitoring protocols and work with users to track tree survival, growth rates, species composition changes, and other vital metrics. A beta version of the web app is available for testing now with mobile app support under development.

TRACKING ENVIRONMENTAL SHIFTS IN TREE BREEDING: UNSUPERVISED MACHINE LEARNING FOR MACROENVIRONMENT CLASSIFICATION

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Tree breeding programs require strategic genetic material allocation to optimize growth and adaptability. Traditional macroenvironment classifications help identify regions with similar environmental conditions, facilitating targeted testing and selection of genetic materials. However, climate change challenges the stability of these classifications, as evolving climatic patterns can render traditional methods less reliable. This study aims to develop a dynamic macroenvironment classification framework to address these challenges. The research was conducted in a 1.6 million hectare Eucalyptus plantation spanning four regions in Brazil. Environmental characterization used 190 environmental covariates (ECs) derived from the SoilGrids database and the Brazilian Daily Weather Gridded Dataset. The study divided the data into historical (2004–2020) and current (2021–2024) datasets to assess environmental changes over time. Unsupervised machine learning, specifically the K-Means clustering algorithm, was employed to classify the macroenvironments. The optimal number of clusters was identified using elbow analysis, determining that 20 clusters best represented the data. Comparing past and current classifications, 50% of the areas showed shifts in environmental classification, demonstrating significant changes in macroenvironmental conditions. Moreover, the emergence of new environmental groups in the current dataset emphasized the dynamic nature of these environments. This new classification approach offers a data-driven method to enhance the strategic planning of field trials and support the adaptation of the current germplasm to evolving conditions. By capturing environmental variability and shifts over time, this methodology can improve the reliability of genetic material allocation and the resilience of breeding programs in the face of climate change.

GENETICALLY DIRECTED DECISION NETWORK FOR ASSISTED MIGRATION OF EASTERN U.S. FOREST SPECIES USING WEB-BASED TOOLS

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Forests provide several ecosystem services throughout the United States and the stability of those services are uncertain with current and future climate change. This causes challenges for forest managers in sourcing climate-adapted seed lots for reforestation and restoration in the eastern United States. This project leverages established web-based tools to streamline the appropriate planting of eastern tree species, including for assisted migration and restoration efforts, by integrating species-specific genetic information into a customized Seed lot Selection Tool (SST). The current SST matches seed lots that are adapted to varying climate scenarios to planting sites. Integrated into the tool are functional traits that establish transfer limits that are based on species genetics. The project will start with Northern Red Oak (*ass*) as a pilot species and expand as data from additional Species with High Restoration Potential (SHRP) become available. This decision network, part of the larger USDA Forest Service funded "Future-proofing Forests through a Genetically Informed Reforestation/Restoration Decision Network" project progresses in three phases: (1) customizing the SST for eastern tree species, (2) mapping eastern nursery locations and production schedules into an online platform, and (3) merging these into a user-friendly interface for forest managers. This ensures that seed sources are optimized for adaptation to future climates, supporting restoration and reforestation efforts.

FUTURE-PROOFING FORESTS THROUGH A GENETICALLY INFORMED REFORESTATION/RESTORATION NETWORK

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Multiple threats to trees and forests, including insect and disease infestation and weather events associated with climate change, will require large-scale tree planting efforts on both Federal and non-Federal lands across the eastern United States. Importantly, broad-scale forest restoration and augmentation efforts will demand an adequate supply of tree seedlings that are genetically adapted to the forests in which they will be planted. Currently, it is unclear which tree species will be the most desirable for forest restoration in the eastern U.S. in coming decades, whether we have sufficient understanding of where to deploy seed sources of those species in light of changing climate conditions, and whether the seed orchard and nursery capacity exists to produce the needed number of seedlings. Our USDA Forest Service-funded project, "Future-proofing forests through a genetically informed reforestation/restoration decision network", aims to close these information gaps by (1) defining and identifying tree species with high restoration potential (SHRP) and assessing their available seed/nursery production capacity and seed transfer guidelines; (2) conducting range-wide genomics studies to refine seed transfer and seed collection guidelines, based on gene offsets, for three common taxa to test the effectiveness of gene offsets for other species that lack common garden studies; and (3) developing a decision network that integrates species-specific genetic information to inform seed collection planning, seed orchard development, nursery production scheduling, and planting site selection to ensure planting activities result in trees with optimal adaptation to future climates. This highly collaborative project encompasses researchers from three universities (Purdue, Penn State, and Kentucky) and Forest Service scientists from the Southern Research Station, Northern Research Station, the National Forest System, and State, Private and Tribal Forestry.

Session 4C: SYMPOSIUM ON MOLECULAR APPPLICATIONS IN TREE BREEDING

GENETICS OF DWARFISM IN *PINUS TAEDA*

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The genetic basis of dwarfism in *Pinus taeda* remains largely unexplored. Dwarf progenies arise from seeds produced on tree branches exhibiting somatic mutations. Half-sib progeny from seed collected from mutant branches segregate for dwarfism. Understanding the genetics of dwarfism in *P. taeda* could elucidate tree height regulation and may have

Presentation Details

Presentation Details

significant implications for breeding programs and mass seed production. To dissect the genetic underpinnings of this trait, we conducted high-density linkage mapping and QTL analysis using open-pollinated progeny from eight mutant families. The seed for five families came from mutant branches of five trees across North Carolina. The seed for the rest of families were collected from 2nd-generation dwarf trees originated from a single mutant branch in Virginia. Phenotypic assessments revealed significant differences between normal and dwarf seedlings in diameter, height, branch number, and branching patterns. These differences were further supported by physiological assessments, including nutrient composition, cone morphology, hormone profiling, and microscopic tissue analysis, suggesting a genetically controlled growth disorder. Seedling progeny from all families displayed approximately a 1:1 Mendelian segregation for dwarfism, indicating the presence of either a major effect locus or a small number of genes, suggesting oligogenic inheritance. Needle tissue was genotyped using the Pita50K SNP array, and linkage maps were generated using a pseudo-testcross approach. Given the challenges of mapping in open-pollinated progeny, this strategy leveraged segregation patterns within maternal half-sib families. QTL analysis identified multiple loci, with the strongest signal on Linkage Group 8. This study establishes the first genetic link between dwarfism in *P. taeda* and mutant branch-derived progeny, identifying key genomic loci associated with the trait. These findings offer valuable targets for marker-assisted selection and functional validation. By linking genetic and physiological mechanisms, we aim to develop dwarf rootstocks that could improve efficiency and safety in seed orchards.

GENE STACKING FOR FUSIFORM RUST RESISTANCE IN *PINUS TAEDA*

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Two major quantitative trait loci (QTL) for resistance to fusiform rust disease, caused by *Cronartium quercuum* f. sp. *fusiforme*, were recently identified in a *Pinus taeda* pedigree (Lauer & Isik, 2021). These QTL are linked in repulsion on linkage group 2, and host recombinant haplotypes inheriting both QTL exhibited near immunity under artificial inoculation. Validation in an independent six-year-old clonal trial confirmed their effectiveness. To assess field performance, trees from the mapping population were planted in two disease-prone sites in Georgia and South Carolina. At planting, all trees carried at least one resistance allele (Rr/rr, rr/Rr, or Rr/Rr) and were symptom-free. After four years, only 3.8% of double heterozygous resistant (Rr/Rr) trees were infected, compared to 50% infection in wild-type (rr/rr) trees, demonstrating the durability of resistance under natural conditions. Full-sib breeding is now underway to combine the two QTL into double homozygous clones for seed orchards, with the goal of producing nearly immune progeny. To facilitate marker-assisted selection, low-cost SNP assays were developed using PACE (PCR Allele Competitive Extension) technology. The SNP markers linked to the resistance QTL were designed and tested against a high-density SNP array using samples from the QTL discovery population, achieving 85–90% correspondence. This study represents one of the first applications of marker-assisted selection in forest trees, providing a model for integrating genomic tools into tree breeding programs.

Presentation Details

GENOME CHARACTERIZATION AND POPULATION GENETIC ANALYSIS OF *CRONARTIUM QUERCUUM* F.SP. *FUSIFORME* IN A CONTROLLED INOCULATION TRIAL

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Fusiform rust, caused by the fungal pathogen *Cronartium quercuum* f. sp. *fusiforme* (*Cqf*), poses a significant economic threat to planted southern pines in the southeastern U.S. Understanding the genetic variability of *Cqf* and its influence on infection rates in southern pines is crucial for developing effective disease management strategies. This study integrates pathogen genome characterization and population genetic analyses to investigate *Cqf* diversity and pine host resistance in a controlled inoculation trial. A total of 5,000 loblolly pine seedlings from five families were inoculated with ten distinct *Cqf* inoculum sources and monitored for infection to explore population-level variation in *Cqf*, we generated an improved genome (86.88 Mbp, 11,996 predicted genes) with high completeness (BUSCO: 91.1%) and structural continuity (Scaffold N50: 12 Mbp). We leveraged this reference for population-genetic analysis of *Cqf*. Population structure analyses revealed an east-west gradient among fungal isolates, independent of the host family, with admixture analysis suggesting a hybridization zone in Alabama and northwest Florida. Analysis of molecular variance (AMOVA) indicated that 83.2% of genetic variation occurred within fungal samples, while 11.5% was attributed to regional differentiation. Infection rates varied by host family and inoculum source, with the most susceptible family ranging from 10.0% to 45.3% and the most resistant family ranging from 1.0% to 20.2%. ANOVA results confirmed that host family, fungal inoculum source, and family-by-inoculum interactions significantly affected host infection rates. These findings highlight the role of regional genetic differentiation and hybridization in shaping *Cqf* adaptability and pathogenicity, providing critical insights informing planting strategies across the southeastern U.S. and for breeding rust-resistant southern pines.

MAJOR QTL CONFER RACE-NONSPECIFIC RESISTANCE IN THE CO-EVOLVED *CRONARTIUM QUERCUUM F. SP. FUSIFORME* – PINUS TAEDA PATHOSYSTEM

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Presentation Details

The endemic fungus Cronartium quercuum f. sp. fusiforme (fusiform rust) causes the most damaging disease of pine species in the southeastern United States. Two bi-parental mapping populations, each consisting of about 1,000 progeny, were challenged with a complex inoculum of 150 pathogen isolates. High-density linkage mapping identified three major-effect QTLs distributed across two linkage groups. All three QTLs were validated using a population of 2,057 cloned pine genotypes in a 6-year-old multi-environmental field trial. In addition to the QTL mapping approach, bulked segregant RNAseq analysis revealed a small number of candidate nucleotide-binding leucine-rich repeat genes containing SNPs associated with disease resistance. The results of this study demonstrate that in Pinus taeda, a limited number of major QTLs can provide effective resistance against genetically diverse mixtures of an endemic pathogen. These QTLs vary in their effect on disease liability and exhibit additivity when combined. When two QTLs on LG2 are inherited together (recombinant haplotypes), the progeny are nearly immune to the rust disease.

Poster Abstracts

EVIDENCE FOR ADAPTATION TO CLIMATE IN LEAF PHENOLOGY TIMING OF NORTHERN RED OAK (*QUERCUS RUBRA*) POPULATIONS

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For many temperate forest trees, spring and fall leaf phenology have evolved in response to local climatic conditions. These timed events balance a trade-off that maximize growth while avoiding the cost of late spring or early fall frosts. However, in a rapidly changing climate, there is increasing potential for mismatch in climatic cues that drive the timing of phenological events. In the context of assisted migration, understanding the strength of local adaptation within and among species to historic climatic conditions will help predict how these relationships may change in the future. *Quercus rubra* is a wide-ranging ecologically and economically important species which may benefit from the implementation of assisted migration. Observations from a provenance trial including 31 populations established between 1962 and 1964 were used to assess relationships between home climate and the timing of spring and fall leaf phenology events. Observations were collected for all populations at three test sites, including average date of bud burst in the spring, average degree of autumn coloration on a given date and average date of leaf coloration, death, and drop in the autumn. Preliminary analysis identified (1) clines in spring bud-burst timing indicating populations with more pronounced continentality flushed earlier than populations with less pronounced continentality, (2) clines in autumn coloration timing, indicating populations from climates with colder autumns turned color earlier than populations from climates with warmer autumns, and populations from drier climates turned color earlier than populations from wetter climates and (3) clines in the duration of the senescence process

indicating populations from warmer climates experience a longer senescence process than populations from colder climates. These results suggest that there is a genetic component to both fall and spring leaf phenological timing and will inform best practices for climate-smart forest management.

PHOTOSYNTHETIC EFFICIENCY AND GAS EXCHANGE DYNAMICS IN GENETICALLY MODIFIED POPLAR VARIANTS

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Short rotation woody crops such as poplar (*Populus* spp.) trees are promising for commercial plantation, biomass and biofuel production, pulping feedstocks, and carbon sequestration. To assess the physiological performance of gene-edited poplar variants with reduced wood lignin ratio, *in vitro* propagation and gas exchange measurements were performed. We propagated and evaluated 17 gene-edited *Populus trichocarpa* variants, with two clones for each variant and a wild-type used as a control, were evaluated in the study. After the in vitro propagation, and later, individual tree plants were moved to the soil for acclimatization in the greenhouse. Before performing leaf gas exchange measurements in the growth chamber with controlled daylight length and temperature. Light response, CO_2 response (A-Ci curves), and dark respiration were measured to evaluate photosynthetic performance under different controlled conditions. A temperature gradient (15–40°C) was used to measure CO_2 concentrations (50–1710 ppm) and light intensities (50–2000 µmol $m^{-2}s^{-1}$). The Farquhar-Berry-von Caemmerer model was opted to estimate the photosynthetic parameters, such as the maximum electron transport rate (Jmax@25) and maximum carboxylation velocity (Vcmax@25). We use statistical analysis to assess the trends of leaf gas exchange among-variant physiological and respiratory responses. This study helps to optimize growth efficiency and environmental resilience for sustainable biomass and pulping feedstock production.

CLIMATE GRADIENTS PREDICT DIFFERENTIATION IN SAMARA AND SEEDLING TRAITS CRITICAL TO GERMINATION FOR *FRAXINUS NIGRA*

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Black ash (*Fraxinus nigra*) is an ecologically and culturally significant keystone species associated with palustrine environments from North Dakota to Newfoundland. Given this broad distribution, understanding how climate gradients impact traits important to adaptation will be critical, particularly in the context of defining guidelines for restoration

and climate-assisted seed transfer. Here, we quantify variation in samara morphology for ex-situ collections collected from across the species' range and relate it to traits associated with emergence success measured in a common garden experiment. We coordinated collection of seed lots from 694 maternal trees representing 75 populations from existing ex-situ collections across the species range. Fifty seeds from each maternal family were x-rayed to assess samara morphology. X-ray images were first segmented into the wing and seed portions of the samara using a machine learning algorithm and then ImageJ was used to calculate area, length, and width of segmented images. In addition to x-ray imaging, a subset of the seeds from 442 maternal families were stratified and planted in the greenhouse to assess early life history variation in emergence success. To test if maternal climate predicts samara morphology, we regressed climate associated with year of origin with morphological traits. We observed a positive relationship between growing degree days (DD>5) and samara traits indicating larger samara were associated with more growing degree days above 5C. This suggests that a longer growing season has a positive impact on resources allocated during fruition. A positive relationship between seed weight and the proportion of emerged seedings per maternal family indicated that greater resources allocated to seed tissue resulted in increased germination success. With predicted temperature increases across North America, our findings suggest that seed lots from maternal populations in regions with longer growing seasons could be valuable to increased emergence and probability of early-life establishment.

RELATING TREE SIZE TO NUTRIENT FORAGING: A STUDY OF APPLE ROOTSTOCK GENOTYPES

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High-density orchards have been facilitated with genotypes that reduce apple tree size. Smaller-sized apple trees (e.g., dwarf trees) are created by grafting compatible genotypes of two or more *Malus* species into a single unit, combining dwarfing belowground genetics (the rootstock) with desirable aboveground varieties (the scion). Presumably, dwarf trees require less soil nutrients than non-dwarfing trees to support tree growth. However, a knowledge gap exists regarding root nutrient acquisition strategies for rootstocks and their relationship to the aboveground phenotype. Established on June 12th, 2024, the NC-140 apple rootstock trial at the Penn State Russell E. Larson research farm presents a unique opportunity to understand the relationship between root foraging plasticity and scion traits. The trial contained two randomized incomplete block designs of 9 rootstocks with two scion varieties ('Aztec Fuji' and, 'Honeycrisp'). We established a root-growth bag experiment to compare root foraging responses by allowing tree roots to selectively grow in a nutrient patch amended with fertilizer (Osmocote 19-6-12) or unamended soil. Root foraging responses are being quantified with estimates of root proliferation and morphological root traits (e.g., diameter). We hypothesized that apple trees grafted on rootstocks that impart larger scion sizes would have: 1) higher root foraging precision, and 2) root traits consistent with an acquisitive strategy (i.e., more proliferation and smaller diameter). Historical data from the NC-140 shows a range for tree cross sectional area (TCSA) between 11.8 to 28.6 cm^3. Estimated with absorptive

root mass, our preliminary results suggest the rootstock with the smallest TCSA (M.9T337) has 78% greater foraging precision than the rootstock with a larger TCSA (V6; p=0.014). This early finding contrasts hypothesis 1, but it does indicate a significant relationship between root foraging precision and tree size. Ongoing analyses will further confirm whether this pattern persists across all 9 rootstocks.

MOLECULAR AND PHYSIOLOGICAL MECHANISMS OF DROUGHT TOLERANCE IN CONTRASTING *Populus Deltoides* GENOTYPES

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Climate change has increased the frequency and severity of droughts, threatening tree plantations and ecosystems. Developing drought-tolerant tree species, such as *Populus deltoides*, is crucial for sustainable forestry and biomass production. While many studies have explored the physiological responses of poplars to drought, the molecular mechanisms underlying drought tolerance in *P. deltoides* remain understudied. To address this gap, I propose investigating drought responses in *P. deltoides* clones with contrasting biomass productivity (high vs. low). Using RNA sequencing (RNA-seq), I will analyze the molecular responses of six genotypes subjected to different drought treatments, with a focus on identifying genes involved in drought recovery pathways. I hypothesize that high-biomass clones will exhibit a delayed but more extensive transcriptional response, with more differentially expressed genes (DEGs) associated with recovery and growth restoration pathways, compared to low-biomass clones, which prioritize survival and may show an earlier but less extensive response. Gene ontology (GO) analysis will be performed to categorize the biological processes, cellular components, and molecular functions associated with the DEGs. Additionally, pathways related to abscisic acid (ABA) signaling, reactive oxygen species (ROS) regulation, stomatal behavior, and biomass growth will be mapped using the KEGG PATHWAY database. By identifying DEGs and their roles in drought tolerance, this study aims to enhance our understanding of the molecular responses in *P. deltoides* and uncover novel genes associated with drought tolerance and biomass productivity. To validate these findings, I will employ qRT-PCR to quantify the expression of key drought-responsive genes. Physiological, morphological, and biochemical data will also be correlated with gene expression results. These findings could inform future breeding programs targeting drought-resilient *Populus* hybrids and contribute to the development of drought-resistant cultivars for commercial forestry.

DIFFERENT CLIMATE SCENARIOS

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Presentation Details

Forest plantations provide vital resources and products for all society. Climate-change scenarios expose forests to environmental hazards, resulting in tree mortality or decreased productivity. Recommending the right species to be planted in the right place is essential and tree breeding programs offer valuable data on species adaptability through field trials. However, even with this information, there is a lack of studies that can provide decision-support models capable of predicting the impact of climate change in site-species recommendations over different environments. This study aims to develop multi-country decision-support models for pine species that can assist in genetic material allocation under current and future climate scenarios. The dataset was composed of 553 genetic trials covering 12 different species of pine, including hybrids, distributed across 12 different countries. The variable selected to express growth potential was the dominant height at age 8 years (HT8). Environmental covariates were obtained from WorldClim 2.1. Random Forest models were fitted for each genetic material and were used to predict HT8 values, generating maps of optimal species allocation. Model evaluation metrics were performed using R-squared (R^2); Root Mean Square Error (RMSE); Mean Absolute Error (MAE). The RF models showed high accuracy, with a mean R^2 of 0.78, MAE of 6.4%, and RMSE of 8.6% across all species. The most widely allocated pure species across both scenarios were *Pinus maximinoi*, *Pinus tecunumanii* high elevation, and *Pinus tecunumanii* low elevation, covering 28%, 16.9%, and 4.2% of the total area, respectively. Under the future scenario, the ranking of species remained consistent, though the proportions shift slightly. The proposed methodology delivers practical models to assist companies to select the best Pine species suitable to their sites. Further research will refine the models by adding tree breeding trial and inventory data from companies and incorporating more environmental variables from other sources.

TRACKING ENVIRONMENTAL SHIFTS IN TREE BREEDING: UNSUPERVISED MACHINE LEARNING FOR MACROENVIRONMENT CLASSIFICATION

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Tree breeding programs require strategic genetic material allocation to optimize growth and adaptability. Traditional macroenvironment classifications help identify regions with similar environmental conditions, facilitating targeted testing and selection of genetic materials. However, climate change challenges the stability of these classifications, as evolving climatic patterns can render traditional methods less reliable. This study aims to develop a dynamic macroenvironment classification framework to address these challenges. The research was conducted in a 1.6 million hectare Eucalyptus plantation spanning four regions in Brazil. Environmental characterization used 190 environmental covariates (ECs) derived from the SoilGrids database and the Brazilian Daily Weather Gridded Dataset. The study divided the data into historical (2004–2020) and current

(2021–2024) datasets to assess environmental changes over time. Unsupervised machine learning, specifically the K-Means clustering algorithm, was employed to classify the macroenvironments. The optimal number of clusters was identified using elbow analysis, determining that 20 clusters best represented the data. Comparing past and current classifications, 50% of the areas showed shifts in environmental classification, demonstrating significant changes in macroenvironmental conditions. Moreover, the emergence of new environmental groups in the current dataset emphasized the dynamic nature of these environments. This new classification approach offers a data-driven method to enhance the strategic planning of field trials and support the adaptation of the current germplasm to evolving conditions. By capturing environmental variability and shifts over time, this methodology can improve the reliability of genetic material allocation and the resilience of breeding programs in the face of climate change.

EVALUATING ENVIRONMENTAL DRIVERS OF SOUTHERN YELLOW PINE NICHE IN THE SOUTHEASTERN U.S. USING SPECIES DISTRIBUTION MODELS

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Southern yellow pine species are critically important components of forest ecosystems across the southeastern United States, contributing significantly to ecological diversity, wildlife habitat, and regional economies. Understanding their geographical distribution, habitat preferences, and environmental determinants is essential for conservation, forest management, and sustainable timber production under changing climatic conditions. This study aims to test multiple species distribution of southern yellow pine species, including loblolly pine (*Pinus taeda*), slash pine (*Pinus elliottii*), shortleaf pine (*Pinus echinata*), and longleaf pine (*Pinus palustris*), across their natural range under future climatic projections. Using geospatial datasets, ecological niche modeling, and spatial analytics, we assessed the primary environmental factors influencing pine distribution, such as climate variables (temperature and precipitation), soil types, elevation gradients, and disturbances (fire frequency and logging intensity). Our results reveal distinct distributional patterns associated with species-specific ecological tolerances and preferences, highlighting significant impacts of climatic variability, soil characteristics, and disturbance regimes on species occurrence and abundance. Particularly, longleaf pine showed strong dependence on fire-adapted ecosystems, while loblolly pine demonstrated broader climatic adaptability. This research provides critical baseline information and predictive insights into potential shifts in southern yellow pine distributions under future environmental scenarios. Consequently, the findings can inform targeted forest management practices, ecological restoration strategies, and policy decisions aimed at preserving these ecologically and economically significant pine species across the southern landscape.

Presentation Details

EXAMINING FOLIAR AND SOIL NUTRIENT CONTENT, SOIL CARBON, AND GENOTYPING OF 12 LONGLEAF PINE GENOTYPES

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Longleaf pine (Pinus palustris) is ecologically important in the Southeastern US. The longevity, high-quality wood, and ability to thrive in diverse environmental conditions make this species resilient and valuable for restoration and tree improvement. Additionally, longleaf pine forests can sequester a significant amount of carbon, functioning effectively as carbon sinks. Despite its ecological and potential economic value, the improvement of longleaf pine has fallen behind species like loblolly pine, resulting in gaps in understanding genotype performance in seed orchards. This study aims to evaluate 12 genotypes in a longleaf pine seed orchard in Eutaw, AL by integrating soil and foliar nutrient analyses, soil carbon fractionation, and genotyping to enhance climate-smart silviculture. Soil and needle samples were collected from 60 trees following standardized protocols and assessed for macronutrients and micronutrients. The soil samples were also analyzed for total carbon, particulate organic carbon (POC), and mineral-associated organic carbon (MAOC) fractions using the Elemental Combustion System (ECS) after sieving and sodium hexametaphosphate dispersion. Genotyping of 43 trees (23 reference genotypes, 20 unknown) will be conducted using the PITA 50K SNP chip to characterize genetic diversity and relatedness. Multivariate statistical models in R will integrate genotype, nutrient, soil carbon, and growth data (height/diameter) to (1) evaluate variations in soil and foliar nutrient dynamics among genotypes, (2) assess genotype-specific patterns in carbon allocation to total carbon, POC, and MAOC fractions, and (3) evaluate the effectiveness of the PITA 50K SNP chip in longleaf pine samples and analyze genetic relatedness and diversity. The study aims to improve longleaf pine seed orchard management strategies to boost nutrient uptake and carbon sequestration, supporting the utilization of enhanced longleaf pine stocks for reforestation and restoration.

ASH PROGENY TAILS RETEST PHENOTYPE REPLICATION IN EMERALD ASH BORER (EAB) EGG TRANSFER BIOASSAYS

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The Emerald Ash Borer (EAB) egg transfer bioassay is used by the US Forest Service Northern Research Station ash breeding program to assess and rank ash trees' resistance to EAB. The bioassay provides a phenotype for each genotype that is tested. When testing clonal replicates of lingering ash selections, typically three grafted ramets of each genotype are tested. Seedlings from controlled crosses are also tested in the bioassay, but they are a

single data point for that genotype. To ensure reliability of selecting the best trees, the most resistant (r tail) and most susceptible (s tail) individuals are propagated for retesting via grafting or softwood cuttings. The grafts and cuttings are bioassayed using the same protocols as the original seedlings. Three variables were used to determine if a ramet's phenotype was an accurate replication of its seedling phenotype: the proportion of host killed larvae, the proportion of fourth instar larvae at eight weeks and the average larval weight. A fourth variable was created to indicate if the ramet agreed for 0, 1, 2, or 3 of the variables. For all variables, family and experiment number had the most significant effect on the reproducibility of the phenotypes. The data suggests that this may be due to the degree of replication within the experiments and families. If there are fewer genotypes or fewer ramets per genotype that were tested within an experiment or family, it tended to show lower reproducibility.

THREE-YEAR SURVIVAL OF CONTAINER-GROWN ROOTED ASH CUTTINGS AND THEIR PERFORMANCE IN EAB (*AGRILUS PLANIPENNIS*) EGG TRANSFER BIOASSAYS COMPARED TO GRAFTS

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The ash breeding program at the US Forest Service Northern Research Station propagates ash trees both to complete replicated potted bioassay experiments in EAB resistance, and to produce material for establishing plantings, including future seed orchards to produce EAB resistant seed. Grafting is successful in ash but leaves some amount of exposed root stock that does not have the same EAB resistance as the scion. Replicating ash using softwood cuttings allows the production of ash on their own roots, which has likely advantages for long-term plantings such as seed orchards. Softwood cuttings of several *Fraxinus* species were rooted in the summers of 2019 and 2020 then grown in containers until the summer of 2023. Over their first winter, 90% of 2019 cuttings and 94% of 2020 cuttings survived. The three-year survival rate of those that survived their first winter was 72% for 2019 cuttings and 70% for 2020 cuttings. They grew similarly to seedlings of the same age and maintained apical dominance with little pruning necessary. In the summer of 2023, 52 of these cuttings were tested in EAB egg transfer bioassays against grafted counterparts to determine if propagation type has a significant effect on a genotype's outcome. Analysis showed that there was no significant effect on either the proportion of tree-killed larvae or the proportion of live fourth-instar larvae. However, propagation type did significantly affect mean larval weight, with larger larvae found more often in cuttings. These results support the continued use of both grafts and cuttings in EAB egg transfer bioassays as well as the validity of past bioassay results.

Presentation Details

THE FIRST GENOME FOR *SASSAFRAS ALBIDUM*: FOUNDATIONAL GENOMIC RESOURCES FOR A TETRAPLOID TREE IMPERILED BY LAUREL WILT DISEASE

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Sassafras albidum, a dioecious tree native to eastern North American forests, faces a severe threat from the invasive redbay ambrosia beetle (*Xyleborus glabratus*), the vector of the laurel wilt pathogen (*Raffaelea lauricola*). This vascular wilt has caused catastrophic mortality to members of the Lauraceae family in the United States, including *S. albidum*, with infected stands experiencing up to 95% mortality within a year. The rapid spread and high mortality rate of laurel wilt presents an urgent conservation challenge for *S. albidum*, particularly in coastal regions. Cytological studies previously reported a somatic chromosome number of 48 for *S. albidum*, whereas most Lauraceae species have 24 somatic chromosomes. Our subsequent flow cytometry and k-mer analyses suggest that *S. albidum* is a cryptic tetraploid, sharing a haploid chromosome number of 12 with many close relatives in Lauraceae. To support population and conservation genetics research avenues for *S. albidum*, we undertook reference genome sequencing. However, accurately phasing haplotypes in polyploid genomes remains a significant challenge. To address this, we utilized PacBio High-Fidelity (HiFi) reads and high-throughput chromatin conformation capture (Hi-C) reads to assemble the tetraploid genome of *S. albidum*, consisting of 48 chromosome-scale scaffolds with an assembly size of 3.4 Gb. Synteny analysis indicates widespread, large structural variation between haplotypes, which is more common in polyploid taxa. Genome completeness was estimated at 98.6% based on BUSCO analysis,and genome annotation identified 23,295 gene models with a protein BUSCO completeness of 93.9%. Additionally, we assembled complete chloroplast (152,539 bp) and mitochondrial (875,944 bp) genomes. These genomic resources provide a critical foundation for future studies on laurel wilt resistance, genetic diversity, and conservation efforts for *S. albidum* and the broader Lauraceae family.

THE LINGERING HEMLOCK SEARCH PROJECT: WE NEED YOUR HELP TO RESTORE THE MIGHTLY HEMLOCK

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To date, eastern hemlocks have displayed little resistance to hemlock wooly adelgid (HWA) at the population level. The majority of eastern hemlock is susceptible to damage from HWA and therefore likely to decline and eventually die once infested with HWA. Breeding HWA- resistant hemlock populations of hemlock is a critical piece of an overall integrated pest management strategy to save eastern hemlock trees from the plight of HWA. In order for researchers to breed native HWA-resistant hemlocks, surviving, or "lingering," trees that may harbor a small amount of genetic resistance need to be found among wild populations. Then, these trees can be tested in a controlled setting and, if found to demonstrate some resistance, bred to increase the level and frequency of resistance. In order for researchers to breed a resistant hemlock, it is important to identify lingering hemlocks for additional evaluation and genetic research. This is where tools like the Lingering Hemlock Search protocol (developed as part of the Nature Conservancy's Tree Species in Peril initiative) and the TreeSnap application come in. We are sending out a call to anyone who is in the forest to be a part of this effort by helping us find and record these lingering hemlocks

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MOBILE AND WEB APPLICATIONS TO ENGAGE THE PUBLIC IN FOREST HEALTH MONITORING

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Forest health monitoring through mobile and web applications is an effective strategy to support forestry research, outreach, and Extension efforts. Our team has developed three applications —**HealthyWoods**, **Eastern Forest Pests**, and **FlorestaDB**—that play distinct but complementary roles in supporting forest health research and management. (1) **HealthyWoods** is a mobile app that empowers forest owners to conduct guided surveys assessing various aspects of forest health, such as regeneration, invasive species, and pathogens. Based on user responses, the app generates a health rating for the surveyed site and directs users to relevant Extension resources for further assistance. Through its web platform, scientists can aggregate and analyze survey data, offering a comprehensive overview of regional forest health trends based on user input. (2) **Eastern Forest Pests** is a reference website focusing on the major pests and pathogens of eastern coniferous and deciduous trees. Initially published as a comprehensive book and website, the Eastern Forest Pests content is now integrated into the *HealthyWoods* app, providing users with educational materials and pest management tools within the same platform. (3) **FlorestaDB** manages large scale, professional forest tree research collections and provides a powerful set of tools to support research from the field to the final data analysis. The mobile and web tools track sample collection, including the chain of custody as well as project management, including details of expeditions, users, shipping, and inventory. Support for wood anatomy, isotope, DNA and mass spectrometry analysis is under development. App features include barcode scanning, GPS tracking, and multi-user collaboration. The current primary use case of FlorestaDB is creating a reference database of international tree samples to combat timber fraud. These applications represent a concerted effort to develop software support that improves scientific research in forestry, offering innovative tools for data and sample collection, education, and forest health monitoring.

Presentation Details

COMPARATIVE TRANSCRIPTOMIC ANALYSES OF DUTCH ELM DISEASE RESISTANCE AND SUSCEPTIBILITY IN DIFFERENTIAL AMERICAN ELM CULTIVARS

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American elm (*Ulmus americana*) is an integral species across forests of North America. Historically, the species was widespread, however its abundance has been severely limited by the arrival of Dutch elm disease (DED) to North America in the 1930's. Since the introduction of the invasive fungal pathogen, *Ophiostoma ulmi* (*O. novo-ulmi*), breeding programs have focused on identifying resistant varieties to restore these iconic, keystone species that were once dominant in forested, riparian, and urban North American ecosystems. An improved understanding of the early post-infection mechanisms of resistance are essential to streamlining the elm selection and breeding process. To identify these characteristics of resistance, we applied dual RNA-seq to characterize the early defense mechanisms associated with DED resistance and susceptibility. Preliminary results suggest differential carbohydrate utilization by the pathogen in a susceptible genotype and increased reactive oxygen species scavenging activity in two resistant cultivars. Results from this study will not only contribute to the growing understanding behind DED responses but also provide molecular markers for furthering American elm breeding programs.

UNDERSTANDING HEMLOCK DEFENSE: SECONDARY PESTS AND FUTURE DIRECTIONS IN HOST RESISTANCE

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Understanding the interplay of native and invasive pests is crucial for preserving forest health. Eastern hemlock (*Tsuga canadensis*), a foundational conifer in eastern North America, is suffering extensive mortality due to the invasive hemlock woolly adelgid (HWA, *Adelges tsugae*). My undergraduate research examined the role of the native hemlock borer (*Phaenops fulvoguttata*), which targets weakened trees, and found that beetles preferentially fly toward hemlocks with higher levels of HWA damage. This suggests that HWA-induced stress may facilitate secondary pest outbreaks. Now, as a PhD student, I am interested in the physiological and chemical traits that may underlie variation in hemlock resistance to HWA. I am particularly curious about the role of volatile organic compounds, secondary metabolites, and differential gene expression in defense, and how these traits may vary among lingering hemlock trees. I hope to connect with researchers studying hemlock genetics to better understand current approaches and identify areas where chemical ecology and genetics may intersect.

Presentation Details

LINGERING ASH GENOMES TO INFORM THE CONSERVATION BREEDING OF CRITICALLY ENDANGERED ASH TREE SPECIES (*FRAXINUS* SPP.) IN NORTH AMERICA

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Emerald ash borer (EAB; *Agrilus planipennis*) has caused unprecedented decline among ash trees (*Fraxinus* spp.) in North America since its introduction in the mid-1990s. While rare, partially resistant individuals have been identified, these "lingering" individuals represent a small fraction (less than 0.01%) of wild populations. We present near-gapless, lingering ash tree genomes for five species: *Fraxinus americana, F. nigra, F. pennsylvanica,* and *F. quadrangulata*, as well as an insecticide-treated specimen of the recently imperiled *F. latifolia*. We show that assembly with deep-learning-based error-corrected Oxford Nanopore simplex reads and Proximo Hi-C sequencing is an effective strategy for producing highly complete (greater than 99% BUSCO completeness) genomes with minimal gaps in non-model plants. We also present complete organelle genomes for each individual. Our approach leverages cutting-edge improvements in long-read sequencing technologies, assembly algorithms, and scaffolding tools that have rapidly increased the tractability of assembling T2T chromosomes with minimal manual curation. While individual species in this study show largely syntenic haplotypes, interspecific synteny analysis suggests that a substantial amount of variation exists even among closely related taxa. As closely related *Fraxinus* have been recently shown to hybridize in the wild, understanding species genome architecture may become increasingly important for leveraging hybrid potential in *Fraxinus* restoration. Finally, we showcase the utility of these resources to restoration breeding by examining the basis of sex determination in *F. americana*, a member of the dioecious, North American ashes in *Fraxinus* section *Melioides*, which will enable marker development for early sex identification.

GENOMICS HIGHLIGHT AN UNDERESTIMATION OF PHENOLOGY SENSITIVITY TO THE URBAN HEAT ISLAND EFFECT

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The phenological timing of leaf out in temperate forests is a critical transition point each year that alters the global climate system, which in turn, feeds back to plants, driving leaf out to occur nearly three days earlier per decade as temperatures rise. To improve predictions of leaf-out timing, urban heat islands (UHIs) or densely developed areas that are hotter than surrounding undeveloped regions, are often used to approximate warming via space-for-time substitutions (ie. rural-to-urban temperature gradients). However, more than just environment changes along these gradients – urban regions are highly managed systems with limited-to-no within species diversity. We demonstrate here that recent observations that UHI gradients underpredict leaf-out response to temperature when compared to temperature gradients through time is likely because both genetics and environment are changing across rural-to-urban gradients, whereas only environment is changing through time. We tested this hypothesis using genomic, phenological, and temperature data of northern red oak (*Q. rubra*) over several years between an urban and rural site. Across our gradient, models that included just temperature predicted moderate advancement of leaf out. However, if we account for the genetic diversity of our trees in our model, leaf out phenology is predicted to advance significantly more in response to temperature. We demonstrate that this stronger relationship between phenological timing and climate is because urban trees have reduced genetic diversity as they are planted from limited stock by humans and, moreover, are most closely related to individuals at the rural site that leaf out later on average.

UNDERSTANDING RANGE-WIDE GENETIC STRUCTURE OF JUGLANS NIGRA (BLACK WALNUT).

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With increasing abiotic and biotic threats to Eastern North American forests there is a need to develop eUective restoration strategies to ensure the maintenance of forest ecosystem function. Climate Smart Forestry provides one approach and emphasizes climate adaptation as a mechanism to inform seed sourcing and seed transfer decisions associated with assisted population migration (APM). Assisted population migration relies on an understanding of the scale and extent of local adaptation to recommend intentional movement of seed sources to mitigate the fitness consequences of rapid climate change. This includes quantifying the contemporary relationship between standing genetic and environmental variation to predict response to projected

environmental change. Quantifying neutral and adaptive genetic variation for forest tree populations can provide an important first step to informing seed sourcing, seed selection and climate-informed seed transfer for reforestation initiatives. Juglans nigra (black walnut), a broadleaf species native to Eastern North America, has been proposed for climate-informed restoration initiatives due to its ability to grow relatively fast, sequester carbon, and provide habitat and food for wildlife. Using a common garden experiment established in State College, PA for a range-wide collection of black walnut provenances, we used ddRad-sequencing to evaluate population genetic structure of black walnut populations sourced from across its range. Long-term we will associate genetic variation with environmental variation and phenotypic variation for traits important to adaptation measured in the common garden experiment. Ultimately, we intend to use genotype environment and genotype-phenotype associations to make climate-informed seed-transfer recommendations, which will facilitate forest tree conservation and restoration in the future.

ASSISTED MIGRATION IS PLAUSIBLE FOR A BOREAL TREE SPECIES UNDER CLIMATE CHANGE: A QUANTITATIVE AND POPULATION GENETICS STUDY OF TREMBLING ASPEN

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A novel method was tested for improving tree breeding strategies that integrate quantitative and population genetics based on range-wide reciprocal transplant experiments. Five reciprocal common garden tests of *Populus tremuloides* were investigated including 6450 trees across western Canada focusing on adaptation traits and growth. Both genetic parameters and home-site transplant models were evaluated. We found a genetic trade-off between growth and early spring leaf flush and late fall senescence. Coefficients of phenotypic variation (*CVp*) of cell lysis (CL), a measure of freezing injury, shrank from 0.28 to 0.10 during acclimation in the fall, and the *CVp* slope versus the freezing temperature was significantly different from zero ($R^2 = 0.33$, $p = .02$). There was more between-population genetic variation in fall phenology than in spring leaf phenology. We suggest that *P. tremuloides* demonstrated a discrepancy between the ecological optimum and the physiological optimum minimum winter temperature. The sub-optimal growing condition of *P. tremuloides* is potentially caused by the warmer ecological optimum than the physiological optimum. Assisted migration and breeding of fast growers to reforest cooler plantation sites can improve productivity. Transferring the study populations to less than 4°C of extreme minimum temperature appears safe for reforestation aligning with the historical recolonization direction of the species. This is equivalent to a 5–10° latitudinal northward movement. Fall frost hardiness is an effective criterion for family selection in the range tested in this study.

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GENOMIC SELECTION ACROSS GENERATIONS OF CLONALLY PROPAGATED NORWAY SPRUCE – EVALUATED ON REAL AND SIMULATED DATA

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The genetic gain from forest tree breeding is hindered by long rotation times due to a combination of late reproductive maturity and the need for rigorous field testing. Genomic selection (GS) aims to accelerate this process by enabling genome wide, marker-based selection in populations without phenotypic data. However, the accuracy of of GS must first be thoroughly tested using data from two generations that has been both phenotyped and genotyped. In this study, we evaluated prediction accuracy across two linked generations of clonally propagated Norway spruce (*Picea abies*): one consisting of 954 plus trees (G0) and 956 progeny trees representing 34 full-sib families (G1). These were tested across a total of 16 field trials across mid- and south Sweden. Both generations were measured for height and genotyped using a 50K SNP chip array. We estimated prediction accuracy both across and within generations, considering between- and within-family accuracy. Various cross-validation scenarios were designed to assess the impact of relatedness between training and validation sets on prediction accuracy. Additionally, we used computer simulations with AlphaSimR to emulate the real data and repeat the same analysis across different historical effective population sizes. Cross-generational GS exhibited considerably lower accuracy than within-generation GS, both in real data (0.15–0.29 vs. 0.49–0.76) and simulations (0.18–0.30 vs. 0.59–0.80). Among cross-generational scenarios, accuracy tended to increase as the training set became more closely related to the validation set, despite even a tenfold reduction in training set size. A combination of direct parents and "distant relatives" provided the most robust prediction accuracy across all simulations. Within-family accuracy was also lower in cross-generational selection. Interestingly, in models incorporating larger training sets with additional unrelated trees, we observed the opposite trend where within-family accuracy consistently improved.

GENOMICS OF WESTERN REDCEDAR: IMPROVING GENETIC RESILIENCE FOR UNCERTAIN FUTURES

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Western redcedar (WRC; *Thuja plicata*) is an ecologically, culturally, and economically important tree species in North America. By creating genomic resources for important trees such as WRC, we aim to develop a better understanding of their biology and improve breeding for growth and resilience to pests and climate change in operational forestry. We present novel transcriptome, genome, and single nucleotide polymorphism (SNP) resources for WRC. Using a genomic selection (GS) approach, we identified thousands of loci putatively associated with growth and specialized chemistry traits in WRC, with substantial variation in effect sizes of loci between different traits. Using a suite of cross-validation approaches, we assessed prediction accuracy in a training population, and validated models for early-expressed foliar traits in a prediction target population of seedlings. Prediction accuracy was high across generations, environments, and ages, and was not reduced to zero among unrelated individuals in the training and target populations, confirming strong linkage disequilibrium and the ability of the model to generate accurate predictions across breeding generations. Genomic breeding value predictions were correlated with those from pedigree but displayed a wider range of within-family variation due to the ability of GS to capture the Mendelian sampling term. Using predicted target population breeding values in multi-trait selection, we functionally implemented and integrated GS into an operational tree-breeding program.

UNRAVELLING GENOMIC DIVERSITY IN THE ALASKAN WILLOW, SALIX ALAXENSIS: A PANGENOME APPROACH TO ADAPTATION IN A RAPIDLY CHANGING ARCTIC

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Relative to a single reference genome, a pangenome provides a comprehensive view of genomic diversity of a species by graphically representing nucleotide and structural variant differences among individuals, including inversions and sequence presence and absence. These genomic differences may also underlie important adaptive differences among populations that would otherwise be largely missed by traditional reference genomes. Here, we present the assembly and annotation of a high-quality reference pangenome for the Alaskan feltleaf willow (*Salix alaxensis*) from long-read sequencing data. The feltleaf willow is a locally adapted and keystone species of Arctic tundra environments and faces one of the most rapidly warming environments on Earth. These and future landscape level samples will be used as part of the NSF-funded Evolving Meta-Ecosystems (EVOME) Biology Integration Institute to understand how rapid climate change influences the evolution and interactions of organisms across interconnected river and tundra ecosystems.

GENETICS OF DWARFISM IN *PINUS TAEDA*

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Presentation Details

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The genetic basis of dwarfism in *Pinus taeda* remains largely unexplored. Dwarf progenies arise from seeds produced on tree branches exhibiting somatic mutations. Half-sib progeny from seed collected from mutant branches segregate for dwarfism. Understanding the genetics of dwarfism in *P. taeda* could elucidate tree height regulation and may have significant implications for breeding programs and mass seed production. To dissect the genetic underpinnings of this trait, we conducted high-density linkage mapping and QTL analysis using open-pollinated progeny from eight mutant families. The seed for five families came from mutant branches of five trees across North Carolina. The seed for the rest of families were collected from 2nd-generation dwarf trees originated from a single mutant branch in Virginia. Phenotypic assessments revealed significant differences between normal and dwarf seedlings in diameter, height, branch number, and branching patterns. These differences were further supported by physiological assessments, including nutrient composition, cone morphology, hormone profiling, and microscopic tissue analysis, suggesting a genetically controlled growth disorder. Seedling progeny from all families displayed approximately a 1:1 Mendelian segregation for dwarfism, indicating the presence of either a major effect locus or a small number of genes, suggesting oligogenic inheritance. Needle tissue was genotyped using the Pita50K SNP array, and linkage maps were generated using a pseudo-testcross approach. Given the challenges of mapping in open-pollinated progeny, this strategy leveraged segregation patterns within maternal half-sib families. QTL analysis identified multiple loci, with the strongest signal on Linkage Group 8. This study establishes the first genetic link between dwarfism in *P. taeda* and mutant branch-derived progeny, identifying key genomic loci associated with the trait. These findings offer valuable targets for marker-assisted selection and functional validation. By linking genetic and physiological mechanisms, we aim to develop dwarf rootstocks that could improve efficiency and safety in seed orchards.

EARLY MULTI-SITE GENETIC EVALUATION OF GROWTH AND PHENOLOGICAL TRAITS IN JUVENILE WHITE OAK (*QUERCUS ALBA*): INSIGHTS INTO HERITABILITY, GENETIC CORRELATIONS, AND ADAPTIVE POTENTIAL

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Plantation forestry is integral to economic sustainability, sustaining employment and industries and maintaining large timberland resources. White Oak (*Quercus alba*) is a keystone species, playing a crucial role as an integral component that maintains the stability of the ecosystem by regulating climate, enhancing biodiversity, and enhancing soil quality, water holding capacity, and wildlife habitat. This study investigates the

genetic parameters of growth and phenological traits in a long-term white oak breeding program conducted across three test sites: Alabama, North Mississippi, and South Mississippi. A total of 3,741 trees from 69 families were evaluated using the ABLUP (pedigree-based Best Linear Unbiased Prediction) method with post-hoc adjustment at multi-environment progeny tests. The model estimated genetic parameters, e.g., narrow-sense heritability, type-B genetic correlations, and variance components for height, root collar diameter, volume, taper, survival, and phenology. Bud break was rated on 0–5 and senescence on 1–6 scales beginning from completely green to leaf senescence. Genetic analyses revealed significant site-dependent variation in heritability, with height (0.94) and volume (0.75) exhibiting higher heritability at North Mississippi site. In contrast, survival and leaf senescence displayed lower heritability (<0.05) in AL and MSN sites. Notably, strong Pearson correlations were observed between height and volume (0.87) for phenotypic variation. Bud break was highly correlated (0.77-0.97) across multiple assessment dates, suggesting consistency in genetic control for this trait over time. The findings emphasized the potential for genetic improvement in white oak through target selection and breeding strategies while a long-term evaluation is necessary for a comprehensive insight into the genetic control and genotype by environment effects of multiple traits for a robust breeding program aiming at advanced productivity and resilience considering the nursery effect.

HIGH-QUALITY RNA EXTRACTION AND REFERENCE GENE EVALUATION IN FRAXINUS

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RNA extraction from woody plant material presents significant challenges due to endogenous phenolics, secondary metabolites, and stem polysaccharides. We have successfully adapted RNA extraction techniques for *Fraxinus* species that rival modern commercial kits. Our approach, tested across multiple tissue types and over 10 *Fraxinus* species, consistently yields high-quality RNA with concentrations exceeding 3000 ng/µL and RIN scores 8.0 - 10.0. This exceptional RNA quality facilitates gene expression analysis focused on low-expressing and regulatory hub genes potentially associated with emerald ash borer (EAB) resistance. We evaluated reference genes as critical quality control standards to establish reliable expression baselines. These reference genes serve as benchmarks for classifying the expression levels of target genes in EAB-susceptible, tolerant, and resistant ash populations. Our work with these recalcitrant species, supports ongoing collaboration with the Chicago Region Tree Initiative to improve urban shade trees, and conserve *Fraxinus* species. The ultimate goal is to enhance the understanding of gene expression patterns across *Fraxinus* populations and identify key genetic markers for EAB susceptibility/resistance. These findings will provide a foundation for developing molecular screening tools for large populations, eliminating the need for lengthy field evaluations and accelerating ash conservation efforts.

Presentation Details

FEMALE STROBILI PRODUCTION OF TOPGRAFTS IN A *PINUS TAEDA* BREEDING ORCHARD OVER ONE DECADE

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Topgrafting is a common technique utilized in loblolly pine (*Pinus taeda* L.) breeding programs to expedite strobilus production to shorten generation intervals and accelerate genetic gain. However, topgraft-induced strobilus production can be quite variable. In an effort to better understand this variability, this study examines the effects of topgraft age (years since topgrafting) on female strobilus production in orchard ramets at the Arrowhead Breeding Center operated by NC State University and the Georgia Forestry Commission. A total of 400 clones were topgrafted into interstock trees between 2014 and 2023, with the majority of clones having scion collected from ortets that were five to seven years from seed, one cohort of older clones (eight or more years from seed), and one cohort of younger clones (two and three years from seed). A common interstock clone was used for all ramets and there was one topgraft clone per ramet. Strobilus production was surveyed annually, and generalized linear mixed models were used to analyze the probability and count of female strobili over time. Results showed that the probability of strobilus production increased with years since topgrafting, but there was considerable year-to-year variation. The percent of topgrafted clones with female strobili one year after topgrafting ranged from 7% to 40%, except for the 2021 season which had 80% of one-year-old topgrafts producing strobili. On average, 55% of topgrafts produced strobili within two years of grafting and 90% within four. The count of strobili produced followed the same trend as the percent of topgrafts producing strobili for a cohort of topgrafts. Clones that were older than 8 years from seed did not tend to produce strobili sooner, but tended to produce more strobili once initiated. Topgrafts from scion that was two and three years from seed produced fewer strobili than expected based on trends from their conventionally-aged counterparts during the first two years after topgrafting, but differences disappeared after the third year. These findings benchmark the effectiveness of topgrafting in reducing generation intervals and highlight the annual variability in female strobilus production. While not predictable, years with prolific strobili production effectively induce reproductive maturity in one-year-old topgrafts.

EARLY RESULTS OF WHITE OAK (*QUERCUS ALBA*) PROGENY TESTS IN MICHIGAN AND WISCONSIN

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Despite its importance, long-term sustainability of white oak is uncertain due to inadequate recruitment of seedlings to replace loss of overstory trees to natural mortality and harvesting. The White Oak Genetics and Tree Improvement Program (WOGTIP) was created to provide a sustainable supply of high quality, improved white oak seedlings to meet current and future demands, improve our ability to conserve and restore white oak in the forest to achieve a variety of ecological, conservation and economic goals at regional and national levels, and to provide genetic resources for academic and industrial research and development. WOGTIP is a collaborative network of 22 regional progeny tests established throughout the geographic range of white oak, and a range-wide provenance test (500 seed sources) in central Kentucky. Of the seed sources in the regional tests, 85% are from seed zones considered locally adapted, 10% are from seed zones just outside of this area, and 5% of the seed sources are from southern latitudes to study climate change responses. Two replicate progeny tests are in Michigan (64 seed sources) and two replicate tests are in Wisconsin (72 seed sources) with a 40% overlap of seed sources between WI and MI. The northern tests in each state are at the edge of white oak's current natural range. Early results show average heights to be similar among the four tests. However, the top performers in each test vary in terms of geographic origin. Local seed zones are generally performing the best in both WI tests and the MI upper peninsula test, but 10% of the seed sources from the southern latitudes are among the top performers in the northern WI and MI lower peninsula tests. G X E interactions and patterns of geographic variation will be detailed in this poster.

HYBRID PROGENY FOR EAB RESISTANCE MARKER VALIDATION

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Fraxinus mandshurica (Manchurian ash) and *Fraxinus nigra* (black ash or brown ash) are geographically distant but genetically similar species that can hybridize and create competent offspring. Manchurian ash is native to northeast Asia and has broad site adaptability similar to that of *Fraxinus pennslyvanica* in North America. Manchurian ash is the co-evolved primary host of *Agilus planipennis* (EAB) and is resistant to EAB when healthy. Black ash is native to northeastern North America, most abundant in swamps, and is highly susceptible to EAB. Two commercially available hybrid cultivars resulting from a *F. nigra* x *F. manshurica* cross, Norther Treasure and Northern Gem, show intermediate resistance to EAB in both common garden studies and EAB egg bioassays. A hybrid breeding program was started to further explore if resistance to EAB from Manchurian ash could be successfully introgressed into black ash. To date we have generated 27 F_1 progeny and 9 back cross progeny. In addition, thirteen hybrid progeny have been shared by the Morden Research Centre, Manitoba, CA. Thirty of these hybrids having been bio-assayed for resistance to EAB. Although this hybrid approach has been largely abandoned in favor of intra-species tree resistance breeding, this hybrid progeny set has shown some value as a validation set for a subset of markers found in *F.*

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mandshurica and *F. platypoda* that are linked to resistance to EAB as found in Kelly et al (2020). We found that four of seven loci show significant linkage disequilibrium in, indicating importance for EAB resistance in *F. manshurica.* We hope to generate a large F_2 hybrid x hybrid cross to better understand the relative importance of each of these loci. These progeny sets can be used to explore other traits of interest in the breeding and deployment of ash resources.

COMPARING FIELD RESISTANCE WITH PINE WILT DISEASE AMONG SIX PINE SPECIES AT SEEDLING STAGES

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Pine wilt disease is caused by the pine wood nematode (*Bursaphelenchus xylophilus*) and has killed many pine trees in Europe and Northeast Asia, including South Korea. In this study, seedlings of six pine species (*Pinus thunbergii*, *P. koraiensis*, *P. densiflora*, *P. parviflora*, *P. rigida* × *P. taeda*, and *P. strobus*) were artificially inoculated by pine wood nematodes in the test field. The Wilt Index was measured every two weeks after inoculation in addition to the mortality rate, detection rate, and pine wood nematode concentration measurement after 24 weeks. The pine wilt disease mortality rates were *P. thunbergii* (80%), *P. koraiensis* (77.8%), *P. densiflora* (62.5%), and *P. parviflora* (22.0%), and both *P. rigida* × *P. taeda* and *P. strobus* survived. The pine nematode detection rates were the same among the species except for *P. rigida* × *P. taeda* (22.2%). High Wilt-Index values were obtained for *P. thunbergii*, *P. koraiensis*, and *P. densiflora*, which had mortality rates higher than the other species. Furthermore, there were no significant differences in the Wilt Indexes between *P. parviflora*, *P. rigida* × *P. taeda*, *P. strobus*, and the control group. Statistically, *P. thunbergii* and *P. koraiensis* showed high susceptibility to pine wilt disease, *P. densiflora* and *P. parviflora* showed moderate susceptibility, and *P. rigida* × *P. taeda* and *P. strobus* showed apparent resistance. These results provide basic data for pine wood nematode resistance breeding or as evidence of the need for afforestation of *P. rigida* × *P. taeda* and *P. strobus*.

IMPROVING SEED GERMINATION AND CUTTED ROOTING OF HONEY BEE SPECIES *(ACER TATARICUM SUBSP. GINNALA)*

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Acer tataricum subsp. *ginnala*, distinguished by its prolific flowering and an average nectar secretion of 0.12±0.01 μL per flower, is utilized as an melliferous species for autumn planting. Although primarily propagated by seeds, natural germination rates are low. Also, the propagation by cuttings is possible, it is under-researched. This study aimed to develop fundamental data for the mass production of quality saplings through the development of both sexual and asexual propagation techniques. In sexual propagation, we compared germination rates with and without cold stratification and removal of the outer seed coat. Removing the seed coat resulted in a fourfold increase in germination rates, achieving a high germination rate of 76.67% after 8 weeks of cold stratification. In asexual propagation, cuttings from 10-year-old trees were treated with 2000 mg/L IBA (Indole-3-Butyric Acid), yielding only an 8% rooting rate. This research highlights the necessity of cold stratification for enhancing germination in *A. tataricum* subsp. *ginnala*. These findings are expected to serve as valuable data for advancing sexual and asexual propagation techniques.

FROM GENES TO POLICY: LEVERAGING GENOMICS FOR CLIMATE ADAPTATION AND ECOSYSTEM RESILIENCE IN CANADA

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Globally, climate change continues to cause severe environmental disturbances, with recent years being the warmest since the beginning of the industrial era. In Canada, the rate of warming is more than twice the global average, with northern regions expected to experience the most extreme changes. This accelerating trend increases the need for scientific guidance on how key species can adapt to climate change, as well as the related economic and societal implications. Genomics provides powerful tools to rapidly assess biodiversity, ecosystem health, and the potential responses of species and ecosystems to environmental change. In response to this need, the Government of Canada launched **GenARCC** (*Genomic Adaptation and Resilience to Climate Change*) in April 2022, through its Genomics Research and Development Initiative. GenARCC brings together

Presentation Details

expertise from across federal departments to inform climate change adaptation and contribute to the protection of Canada's biodiversity, ecosystem resilience, food security, and public health. The initiative uses genomic tools to quantify genetic diversity, monitor ongoing changes, and predict the future vulnerability of species and populations across agricultural, aquatic, coastal, forest, and tundra ecosystems. The forest and tundra component of GenARCC applies a multi-taxa, multi-omic approach, integrating genomic, functional, phenotypic, and ecological data for a wide range of species —including keystone trees, wildlife, associated microbes, and pests. Focal species include black spruce, balsam poplar, spruce budworm, and Septoria. A key objective is to use ecological and genomic modeling to assess species' climate vulnerability and identify geographic hotspots of risk or resilience.This broad, collaborative effort is designed to support climate mitigation strategies and inform policy and regulatory decision-making, providing decision-support tools for a wide range of end-users working to sustain Canada's ecosystems in a rapidly changing climate.

Field Trips

The four track options for the WFGA/NFGA/SFTIC joint meeting have been created to examine the full gamut and application of genetics in forests. Participants can evaluate species diversity and the significance of ex situ germplasm collections for common garden studies, essential for long-term conservation. The tour will cover nursery operations aimed at seedling propagation and reintroduction strategies, emphasizing the importance of restoring native populations. Additionally, discover the ongoing research into the introgression of pest and disease resistance traits, crucial for enhancing forest resilience against invasive species. Engage with experts to deepen your understanding of these vital ecological processes.

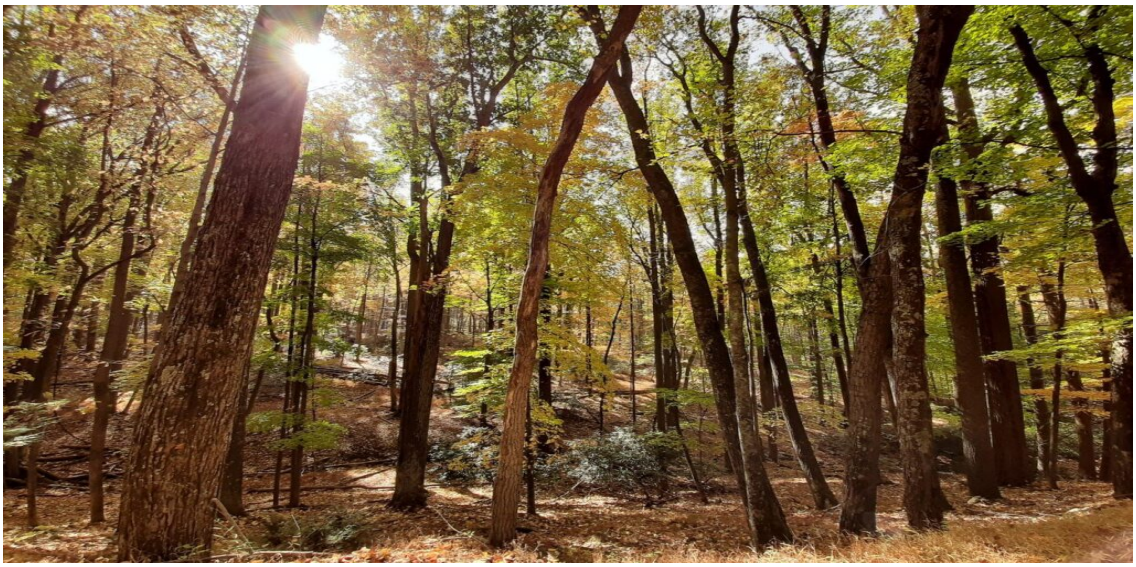
Track 1: Campus

Research groups at Penn State University are working on various species-specific initiatives highlighting the critical role of genetic diversity in ecosystem restoration efforts. The **Black Walnut Provenance Study**, established in 1980, investigates climate change effects on tree populations. Concurrently, **American chestnut trials** focus on germplasm conservation and developing disease resistance through hybrid breeding and genetic engineering. The **"PopUp Poplar" program** features a common garden of 100 poplar clones, highlighting genetic diversity's role in adapting to climatic changes. Additionally, a visit to the devastated **green and white ash** provenance trial alongside new **black ash** plantings reveals insights into the interplay of genetic and environmental factors in climate adaptation and resistance to the **emerald ash borer (EAB)**.

Walking Expectations: Light walking, no more than 250 feet at a time, over slightly uneven but flat terrain can be expected. Trucks will be provided for those who would prefer to ride.



Field Trips



Track 2: Penn Nursery and Rothrock Forest

Two key highlights of this track include a visit to large, treated **eastern hemlock** as well as a disjunctive population of **red spruce** at a fen wetland, a unique case study in genetic adaptation to climate variability. Pennsylvania DCNR's **Mira Lloyd Dock Resource Conservation Center** encompasses **Penn Nursery** (established 1922) and operational aspects of a forest tree nursery including seed cleaning, seedling processing, and sign and picnic table construction operation. Participants will explore the diverse work occurring in the 40+ acres of nursery seedling beds, seed orchards, and provenance trials for species such as **white pine**, **shortleaf pine**, **white oak**, **red oak**, and American chestnut. From seed orchard establishment, seed sourcing and more, this track offers a comprehensive exploration of how genetics informs sustainable forestry practices.

Walking Expectations: At the nursery, light walking, no more than 250 feet at a time, over slightly uneven but flat terrain can be expected. Trucks can be provided at Penn Nursery for those who would prefer to ride. At the red spruce/hemlock stop, a walk along a trail with uneven footing of 0.5 miles will occur. A few smaller red spruce can be viewed from the roadside if mobility is a concern.

Track 3: Shavers Creek/ Stone Valley

Participants will journey to two of Penn State University's premiere facilities- Shaver's Creek Environmental Center and the Stone Valley Experimental Forest-to engage with studies examining species adaptation, resilience, and their interaction with broad-scale abiotic conditions. At Shaver's Creek, tour fenced, common gardens focusing on non-timber forest products (NTFPS), especially that of a newly discovered ramp species. The tour will also include the **Shale Hills Critical Zone Observatory**, where attendees will learn about critical zone studies, Pennsylvania's geology, and climate measurement equipment. Moving on, the group will observe the work of the Kaye Forest Dynamic Lab which has established common garden experiments using assisted migration. The trip will conclude at **Musser Gap**, highlighting restoration efforts and non-timber forest products.

Walking Expectations: Light walking, no more than 250 feet at a time, over crushed gravel walkways with very slight incline. To view climate measurement equipment will require a short walk up a steep incline. A UTV may be available to assist those with mobility concerns.



Field Trips



Track 4: Campus / Penn Nursery Combo

Take a hybrid tour of sites between both the **Penn State campus** and **Penn Nursery**, enjoying slightly condensed versions of the two. This combined track integrates the genetic research conducted on campus with the practical applications at **Penn Nursery**. Participants will explore the genetic diversity of tree species in the Arboretum at Penn State, including **American chestnut** and **poplar**, while learning about nursery practices that enhance genetic conservation. The focus on the **white oak, shortleaf pine,** and additional **ash and American chestnut trials** will illustrate how genetic research informs restoration strategies. This track provides a comprehensive understanding of the intersection between genetics and sustainable forestry, highlighting the importance of genetic diversity in ecosystem health.

Walking Expectations: Light walking, no more than 250 feet at a time, over slightly uneven but flat terrain can be expected. Trucks will be provided for those who would prefer to ride.

Career Panel



Sara Fern Fitzsimmons
Chief Conservation Officer at The American Chestnut Foundation



Dr. Nathalie Isabel
Research Scientist at Laurentian Forestry Centre in Canada



Dr. Mengjun Shu
R&D Associate Staff Member in the Bio-Sciences Division at Oak Ridge National Laboratory



Dr. Meg Staton
Associate Professor at University of Tennessee



Dr. Alan T. Whittemore
Research Botanist at U.S. National Arboretum

Pre-Workshops

TreeSnap/TreeSnapPlots

Organizers: Meg Staton, Florence Caldwell, Chance Stribling – University of Tennessee, Knoxville

Location & Time: 106 Forest Resources Building, Monday, June 23rd 9:00am - 10:00am

Engaging the public in research enhances forest projects by broadening geographical scope, providing more frequent data points, and enabling early detection of forest health issues. To integrate non-professional scientists, an easy-to-use platform is essential for guiding new participants. We have created mobile and web applications to facilitate public science, focusing on two case studies. TreeSnap is a mobile app that encourages the public to scout for trees affected by pests and diseases, such as ash, chestnut, hemlock, and elm. Volunteers can use the free app to submit GPS locations, images, and traits relevant to tree research. We enhance data utility by incorporating customized questions for each species, allowing scientists to sort, filter trees, and access user observations on the TreeSnap website. With over 2,000 active users and over 20,000 tagged trees, TreeSnap aids scientists in identifying trees that may have genetic resistance. We are also developing a new software system for plot monitoring, enabling participants to report on tree survival and health over the years, with a focus on hemlock species threatened by hemlock woolly adelgid. Our six years of experience in creating user-friendly apps for forest research demonstrates that effective software fosters valuable data and participant satisfaction.

Capacity: All Welcome

North American Quantitative Genetics

Organizer: Fikret Isik – NC State University

Location & Time: 107 Forest Resources Building, Monday, June 23rd 1:00-5:00pm

The Forest Genetics Conference 2025 will include a per-conference workshop on quantitative genetics. Attendance is limited to 40 participants, and the organizers reserve the right to cancel the workshop if the minimum number of 15 participants is not met. This workshop will provide a platform for geneticists and breeders to exchange ideas regarding the implications of quantitative genetics in managing forest genetic resources and breeding. Key topics will include using DNA markers for pedigree quality control and a pilot project on genomic prediction involving a multi-generational Loblolly Pine population in the NC State University Tree Improvement Program.

Capacity: 40 people

CartograPlant/TreeGenes

Organizers: Meghan Myles, Brandon Lind, Jill Wegrzyn – University of Connecticut

Location & Time: 107 Forest Resources Building, Monday June 23rd 10:00-12:00pm

Description: Combining well annotated genomics datasets can provide critical insights for plant populations. CartograPlant (**cartograplant.org**) is an open-source, map-based web application that integrates genotype, phenotype, and environmental data from multiple sources. Built on the Tripal platform, CartograPlant allows users to easily filter, analyze, and visualize georeferenced plant populations. Through the Tripal Plant Popgen Submit Pipeline (TPPS), researchers can submit standardized data with metadata following FAIR principles. CartograPlant connects the TreeSnap mobile application to enable field to analysis connections. CartograPlant hosts over 900 environmental layers, supporting complex meta-analyses, including diversity estimates, population structure assessments, and association mapping. Analysis is enabled through high-performance computing and reproducible workflows paired with interactive visuals in NextFlow.

Capacity: All Welcome

Post-Workshops

Oak Pangenome Workshop

Organizer: Dr. John Carlson, Academy Professor and Professor Emeritus of Molecular Genetics, Former Director of The Schatz Center for Tree Molecular Genetics

Location & Time: 217 Forest Resources Building, Friday, June 27th 1:00pm- 3:30pm

The **Oak Pangenome Workshop** will be a meeting of the JGI Community Science-Program project "Super-Pangenomes for gene discovery and climate- resiliency research and breeding in Eastern Oak Synameons. The research platform being developed at JGI will consist of newly sequenced, assembled and annotated genomes and SNP diversity data for 16 oak species, plus SNP data for existing Northern Red Oak (*Quercus rubra*) and White Oak (*Quercus alba*) families. An overview of progress on the pangenome research platform will be presented followed by an open discussion of future research directions and group investigation opportunities enabled by the new platform.

Capacity: All are welcome – Lunch will be provided