



Seed Orchard Conference

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Proceedings

*Seed Orchards - The Key Stage in Forest Tree Breeding and Sustainable Forest
Management*

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TOPICS



1. Seed Orchard Design
2. Genomics Analysis
3. Reproductive Phenology and Seed Production
4. Mating Patterns & Gene Flow Using Molecular Markers
5. Genetic Gain & Diversity of Seed Orchards Crops
6. Effect of Management on Cone Production
7. Relationship of the Seed Orchards with Other Forest Issues

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TABLE OF CONTENTS

TOPIC 1. SEED ORCHARDS DESIGN.....	1
Seed Orchards Under Rolling-Front Landscape Breeding.....	2
Seed Orchard Evolution from Traditional Seed Production to Advanced Genetic Gains and Breeding Base.....	3
Expanding the Belgian Seed Orchard Network, a Collaboration Between Private and Public Sector	4
Ash in Distress: Results from a Large Experimental Field Trial on Ash Dieback in Austria	5
Challenges for a Seed Orchard Programme Towards Seed Availability for Future Forest Composition in Saxony, Germany	6
TOPIC 2. GENOMIC ANALYSIS.....	7
DNA Sequence Data: Transition from Quantitative Genetics to Quantitative Genomics	8
A Genetic Database As a Tool To Provide Proof of Origin of Autochthonous Forest Reproductive Material.....	9
Genomic Prediction and Gwas in Conifer Genetics and Breeding: Foreseeable Challenges and Solutions	10
TOPIC 3. REPRODUCTIVE PHENOLOGY AND SEED PRODUCTION.....	11
Reproductive Phenology, Genetic Diversity and Seed Production in Clonal Seed Orchards.....	12
Transcriptome and Morpho-Physiological Analyses Reveal Factors Regulating Cone Bud Differentiation in Qinghai Spruce (<i>Picea Crassifolia</i> Kom.).....	13
Phenotypic Plasticity of Selected Reproductive Traits of European Larch Clones on Seed Orchards	14
TOPIC 4. MATING PATTERNS & GENE FLOW USING MOLECULAR MARKERS	15
Understanding Mating Dynamics and Mitigating Pollen Contamination in Forest Tree Seed Orchards: Insights and Strategies.....	16
Low Influence of External Pollen Sources in Swedish Norway Spruce Seed Orchards	17
Ensuring Reliability and Quality of New Seed Orchards Along the Track from the Breeding Population to Seedlots in French Varieties of Loblolly Pine and Douglas Fir.....	18
The Contribution of Individual Pollen Donors in a Hybrid Larch Orchard from Northern Germany.....	19
TOPIC 5. GENETIC GAIN & DIVERSITY OF SEED ORCHARDS CROPS	20
Estimation of Genetic Gain and Gene Diversity for Seed Orchard Crops	21
Evaluation of Genetic Diversity and Gain in <i>Eucalyptus Camaldulensis</i> Seed Orchards of Two Generations.....	22
How Do Phenotypes of Seed Orchard (Improved) vs. Wild (Unimproved) Seedlings of <i>Pinus Contorta</i> (Lodgepole Pine) Differ Under Commercial Greenhouse Condition?	23

Variation of Strobili and Cone Productions in Clonal Seed Orchards of Anatolian Black Pine...	24
Advanced - Generation Seed Orchards of European Silver Fir in Romania – Achievements and Perspective.....	25
A Low Input - High Selection Intensity - Approach for Rapid Development of Ash Trees With High Tolerance Towards Ash Dieback Based On Breeding Seed Orchards (BSOs)	26
Enhancing Genetic Improvement Clonal Seed Orchards Of <i>Pinus Densiflora</i> Through Fertility-Based Clone Selection.....	27
Genetic Diversity of Silver Fir and European Larch Seed Orchards in Romania - Support for Advanced Breeding Generations	28
A Novel Allelic-Based Stochastic Simulation Modeling for Seed Orchard Development	29
Genetic Parameters And Genetic Gains from Half-Sib and Full-Sib Progenies of <i>Larix Decidua</i> (Mill) Seed Orchards in Romania.....	30
TOPIC 6. EFFECT OF MANAGEMENT ON CONE PRODUCTION.....	31
Management of the Fourth-Cycle Seed Orchard in Chinese Fir.....	32
High Stand Density Improves Seed Production in Seed Orchards of the Masting Species <i>Picea Abies</i>	33
Conifer Seed Orchards in The US Pacific Northwest	34
Site Variation in Coning and Nutrient Content in Danish Nordmann Fir Seed Orchards	35
Mass-Production of Hybrid Larch Forest Reproductive Material: Is Advanced-Generation Hybridisation Seed Orchards an Option?.....	36
TOPIC 7. RELATIONSHIP OF THE SEED ORCHARDS WITH OTHER FOREST ISSUES.....	37
Unveiling The Spectrum: Analyzing Genetic Parameters of Hyperspectral Reflectance and Leaf Functional Traits in Replicated Clonal Seed Orchards With Annual Dynamics	38
Unveiling Nordmann Fir Susceptibility to Silver Fir Woolly Adelgid: Insights from Clone Seed Orchards.....	39
Scots Pine Seed Orchard Experiment Gives Indications of Pollen-Mediated Transgenerational Epigenetic Inheritance	40

POSTERS

Spring Phenology and Genetic Diversity of <i>Quercus Robur</i> L. (Pedunculate Oak) in Movileni Seed Orchard	42
FORSEE: Seeds for Climate-Adapted Forests: New Strategies for Management of Seed Orchards, Seed Storage and Germination Testing	43
Development of a Non-Destructive Method for Assessing Species Purity in Euro-Mediterranean Firs	44
Advancing Seed Orchard Design through Integrated Arrangement and Evaluation Models.....	45
Seed Orchards are Key to Increasing Resources for the Future	46
The First Grayish Oak Seed Orchard Established in Romania	47
An Overview of Seed Orchards in Ireland	48
Genetic Background of Early Flowering in <i>Betula Pendula</i> for the Successful Development of Efficient and Rapid Breeding	49
Assembly and Analysis of Chloroplast Genome for DNA Marker Development in <i>Quercus Acutissima</i> ‘Gumsura 1ho’	50
The Newest Seed Orchard of <i>Picea Abies Pendula</i> form Established in Romania.....	51
Challenges for Implementing Assisted Migration into a Seed Orchard Programme in Saxony, Germany	52
Study of the Decline of Coniferous Fruiting in the Landes Massif.....	53
Flowering and Seed Production on 20-Year-Old Silver Birch Clone Seed Orchard in Central Poland	54

TOPIC 1.

SEED ORCHARDS DESIGN

KEYNOTE PRESENTATION

SEED ORCHARDS UNDER ROLLING-FRONT LANDSCAPE BREEDING

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In the context of climate change, traditional genetic gain predictions become unreliable. Tree breeding strategies must adapt rapidly to changing environmental conditions, becoming self-regulatory. Rolling Front Landscape Breeding, a new concept originating from the earlier Breeding-without-Breeding approach, replaces conventional breeding cycles with continuous evaluation and selection. The method involves genetic evaluation across forest stands of successive ages and broad ecological gradients, enhancing the adaptive response. Unlike traditional methods, there are no distinct breeding cycles in Rolling Front Landscape Breeding, allowing seed orchards to remain indefinitely in a given location. This is facilitated by dividing the orchard into two tiers: the reservoir and the core. Top-ranking genotypes from a candidate population are grafted into the reservoir until they reach reproductive age, at which point they may enter the core subset. As new stands progressively reach selection age and undergo periodic genetic evaluation, new genotypes become available in the candidate population. This allows for periodic shifting of candidates into the orchard and the possible removal of those that become either genetically inferior or reproductively inactive. My presentation will review this concept in detail, focusing on optimizing seed orchard logistics.

Keywords: climate change, adaptation, gene diversity, tree improvement, genetic evaluation, *in situ* selection

SEED ORCHARD EVOLUTION FROM TRADITIONAL SEED PRODUCTION TO ADVANCED GENETIC GAINS AND BREEDING BASE

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A traditional tree breeding program typically started with collecting phenotypically superior parent trees and grafted them into gene banks, and early on served as seed orchards (unrouged). Following progeny testing, seed orchards may be rouged to include only high genetic value parents for greater genetic gains. This traditional seed orchard operation has been very effective for reforestation in the southern US and generated significant genetic gains in the loblolly pine breeding program for over 60 years. Trees from 1st-generation seed orchards have produced 7-12% more volume at harvest than trees from wild seeds, while trees from the 2nd-generation orchards produced 21% in rotation volume over unimproved trees. When 2nd-generation seed orchards are rouged based on progeny tests, genetic gains can be further increased to 30%. The 3rd-cycle seed orchards are estimated to be about 15 % genetic gains over the 2nd-generation seed orchards. Additionally, these seed orchards are being used for the operational Controlled Mass Pollination with high value genetic parents, which can further increase genetics gain (up to 60%) by increasing the selection intensity, capturing the non-additive genetic variance, and eliminating pollen contamination.

As the new cycles of genetic materials become available, these traditional seed orchards are now serving as the breeding orchards with the top-grafting, e.g., the newly selected juvenile trees from progeny tests with the best genetic values are directly grafted on the top of existing and mature seed orchard trees to produce early flowering. This approach has been successfully used in loblolly pine breeding and significantly reduced the breeding cycle. With the 4th cycle breeding on the way, this top-grafting method is deployed to start 5-th cycle breeding. With the newly developed genomic breeding strategy, the juvenile materials from the genomic selection will be top-grafted onto the mature seed orchard trees to further reduce breeding cycles.

Keywords: gene bank, rouged orchard, controlled mass pollination, top-grafting, breeding orchard

EXPANDING THE BELGIAN SEED ORCHARD NETWORK, A COLLABORATION BETWEEN PRIVATE AND PUBLIC SECTOR

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By 2030, the regional government of Flanders (Belgium) needs to create an additional 10,000 hectares of forests. Due to the slow pace of natural reforestation in non-forested areas, a substantial amount of planting material is required. This high demand for forest reproductive material (FRM) has led to shortages in the existing supply. FRM refers to seeds, plants, and parts of plants that are used for planting new forests and other types of tree planting. FRM is harvested from registered parent trees (i.e., basic material) to ensure traceability. Certification of FRM ensures its origin and quality. To solve the problem of shortages, the Research Institute for Nature and Forest (INBO) collaborated with the Flemish Agency for Nature and Forests (ANB), which owns most of the seed orchards, and the tree nursery sector, and carries out the harvesting and commercialization. We address the challenges of shortages in FRM through three approaches. Firstly, we are analyzing existing seed orchards to identify factors that cause a decrease in seed production. Secondly, we duplicate material, using various methods to expand or rejuvenate the public seed orchards. Lastly, duplicated material from the public seed orchards is being provided to establish private seed orchards. Throughout the project, we have identified numerous flaws in the existing seed orchards, such as light availability and pruning practices; we have identified various shortcomings and areas for improvement in the existing seed orchards, such as light availability, pruning practices, or diseases. Fortunately, many flaws are temporary and can be mitigated by adjusting management practices. The material used for expanding and constructing seed orchards, both private and public, is clonally propagated ensuring uniform quality. Various propagation methods, including softwood and hardwood cuttings, grafting, and rootstock, have been employed with varying degrees of success. Nevertheless, substantial progress has been made in duplicating material during the project's first year.

Keywords: Seed orchard design, Seed orchard duplication, Seed orchard establishment, cuttings, grafting

ASH IN DISTRESS: RESULTS FROM A LARGE EXPERIMENTAL FIELD TRIAL ON ASH DIEBACK IN AUSTRIA

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Common ash (*Fraxinus excelsior*) is a highly valuable broadleaved species whose survival is threatened by the alien invasive ash dieback pathogen *Hymenoscyphus fraxineus*. Studies provided evidence that ash dieback tolerance has a polygenic basis and identified tolerance-associated genetic loci (Stocks et al., 2019). Here, we report on one of the largest extant field trials aiming at the identification and characterization of ash dieback tolerant genotypes, while encompassing the genetic diversity of common ash in Austria. The overall goal of the project “Ash in Distress” is the selection of highly disease-tolerant clones and in consequence the establishment of seed orchards to provide improved reproductive material to forest owners. In detail, seeds from 715 disease-tolerant female trees were collected in 2015 and 2017, giving rise to more than 35,000 offspring which were raised in a common garden. Disease incidence and severity were evaluated for three consecutive years for all seedlings. Eventually, approx. 20% of the evaluated offspring remained disease-free, providing a broad base for further selection. We observed that susceptibility in juveniles can strongly increase during the first three years following germination, whereas trees showing no disease symptoms towards the end of the third vegetation period are likely to remain symptomless or become only slightly damaged over the following years. Genotype-tolerance association analyses using the 4TREE array targeting 13,407 single-nucleotide polymorphisms (SNPs) were performed. Overall, our genotype-tolerance association analyses failed to detect candidate loci that could be used for future genotype-informed breeding. Nevertheless, the array proves powerful in studying genetic structure and detecting introgression from closely related species, as well as exploring associations with environmental variables and other traits of interest.

Keywords: *Fraxinus excelsior*, ash dieback, resistance breeding, seed orchards

CHALLENGES FOR A SEED ORCHARD PROGRAMME TOWARDS SEED AVAILABILITY FOR FUTURE FOREST COMPOSITION IN SAXONY, GERMANY

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Saxony, one of the 16 German federal provinces, is situated in Southeast Germany. It covers relatively different growing regions within a rather small area. The three most important regions refer to the low mountain range (e.g., Ore Mountains) along the southern border with Czechia (medium fertile and cooler, precipitation p $550 \leq 830$ mm), fertile hilly loess areas in the transition zone, and Pleistocene lowlands in basins with fertile soils (loam, silt) or with poor sand soils under more continental climate with $p \approx 500$ mm. The Saxon State Forest enterprise (SBS) is therefore in charge of sustainable forest operations in a large variety of different forest types in the Saxon state-owned forests (205,370 ha, = 39.5 % of the total Saxon forestland; www.wald.sachsen.de, accessed 2024-03-14). However, after the past 30 years of active ecological forest re-structuring, the Norway spruce (*Picea abies*) and Scots pine (*Pinus sylvestris*) remain the main species. This is the consequence of approx. 200 years of anthropogenic shifts in species composition towards efficient wood supply in order to meet the wood demand of mining, construction, and pulp & paper sectors. Today, drastic losses of these softwood stands are caused by the yet-running climatic change with more frequent droughts and storms, followed by dramatic bark beetle calamities. Establishing climate resilient (i.e., genetically diverse) stands with more than only native species or provenances result in an increasing demand for forest reproductive material that cannot be met by approved seed stands alone in the future. As there are no relevant private operations in the creation of forest seed sources (in compliance with the federal act on forest reproductive material FoVG), the SBS takes responsibility for the entire Saxon forests by implementing a 5-years seed orchard programme. It has the objectives of (i) evaluating the existing seed orchards, (ii) stipulation of clear operational responsibilities for the management of seed orchards, (iii) maintaining/complementing existing orchards to achieve the legal (FoVG) approval as well as (iv) the establishment of approx— 6 new seed orchards under given species priorities.

The present contribution will present an overview of existing seed orchards, explain why the majority has been established for coniferous species, and summarise the progress with the first two de novo establishment activities (hornbeam, Norway maple). One central challenge is the selection of plus tree collectives for the new seed orchards that will provide both a genetically diverse seed yield for the best possible stand adaptation capability and genetic gain for maintaining the stem or wood quality. The presentation will illustrate the largely species-dependent approaches that are due to different dispersal and abundance of the species within the administrative regions of provenance.

Keywords: plus tree selection, seed orchard programme, hornbeam, norway maple, saxony

TOPIC 2.

GENOMIC ANALYSIS

KEYNOTE PRESENTATION

DNA SEQUENCE DATA: TRANSITION FROM QUANTITATIVE GENETICS TO QUANTITATIVE GENOMICS

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The availability of DNA sequencing has instigated a paradigm shift, ushering in the era of quantitative genomics. Traditional quantitative genetics relies on the average numerator relationship (A-matrix), providing global estimates for mating designs' half-sib (HS = 0.25) and full-sib (FS = 0.5) family members but overlooking the Mendelian sampling term. This omission hinders the estimation of variation among HS and FS family members. In contrast, DNA sequencing data accurately determines the actual fraction of alleles shared among HS and FS family members, enabling the estimation of their realized genomic pairwise kinship (G-matrix), thus capturing their true similarities and differences. The strength of the G-matrix is facilitated through encompassing both contemporary and ancestral pedigree, a pivotal component for accurately determining the realized pairwise kinship among individuals. Acknowledging the G-matrix role as the cornerstone of subsequent quantitative genomics analyses is crucial, emphasizing the need for its proper formation to ensure accurate determination of genetic parameters. In this context, we will introduce various SNP selection protocols to showcase the benefits of incorporating the "best" SNPs in the G-matrix formation. Furthermore, we will compare the impact of SNP selection on variance components inferences and theoretical accuracy of breeding values in two species, one with a large (white spruce) and the other with a small (Eucalyptus) genome.

Keywords: quantitative genetics, DNA, SNP, genome

A GENETIC DATABASE AS A TOOL TO PROVIDE PROOF OF ORIGIN OF AUTOCHTHONOUS FOREST REPRODUCTIVE MATERIAL

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Under the European Green Deal, the ambitious goal of planting 30 billion extra trees across Europe has sparked interest in (re)forestation projects. However, the origin of planted trees is key for their resilience, resistance and adaptation potential to future climate conditions and should thus be taken in consideration. In Flanders, Northern Belgium, a control system is being established based on molecular markers to verify the origin of Forest Reproductive Material (FRM) certified in the category ‘source identified’.

Across Europe, few countries have already made use of molecular techniques as a method to control the origin of certified FRM. In Flanders we are adopting a unique approach by setting up a genetic database as a tool to verify the origin of FRM. The genetic database will contain genotypes of all parent trees present in our Flemish autochthonous seed orchards of nine economically or ecologically important tree and shrub species. The genotypes are set up with microsatellite markers. For setting up this database, new microsatellite markers were developed for two species.

Primarily, the genetic database will be used for parentage analyses and/or assignment tests to verify the origin of forest plants sold by tree nurseries. However, the database has several more applications. Genetically mapping of the autochthonous seed orchards enables us to evaluate the genetic diversity within the orchards and identify any presence of hybrids or cultivars. Knowing the exact locations of the genotypes within a seed orchard also aids in establishing new copies of the seed orchard. Moreover, the genetic database and microsatellite markers will be made publicly accessible, enabling cross-border comparison of genetic diversity between different stands. The genetic database will thus not only be a tool to control the origin of the FRM, but also aid seed orchard management and ex situ conservation of genetic diversity.

Keywords: seed orchard, forest reproductive material, microsatellite markers, genetic conservation, genetic diversity

GENOMIC PREDICTION AND GWAS IN CONIFER GENETICS AND BREEDING: FORESEEABLE CHALLENGES AND SOLUTIONS

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Genomic prediction (GP), where phenotype predicted from genetic variants, is driven modern plant and animal breeding, resources management, and human medicine. Genome-wide association study (GWAS), where genetic dissection enabled by genotype to phenotype prediction, is critical for genetic understanding of phenotype variation. Currently, GP and GWAS are seen related but different, one may need several of thousands of SNP sites, and the other may need all measurable variants from the whole genome. GP and GWAS computation are well established in some way. However, challenges may come with the fast advancing sequencing, phenotyping techniques and climate data collection resolution, especially for conifers which have huge genome sizes, unprecedented amount of variant, and genetic (reflected in potentially complex genetic interaction) and environmentally (multi-year effect) complex phenotype.

(1) Traditionally, the linear equation $P = G + E$ has modeled the genetic (G) and environmental (E) contributions to phenotypic traits (P), assuming they contribute independently to a given phenotype. Should we consider incorporating $G \times E$ interactions?

(2) The environment has typically been reduced to a few grouped and representative variables. Should we aim to include every single real environmental variable and their interactions with each genetic variant?

(3) Although thousands of SNP sites are currently sufficient for genomic prediction, can we envision a future where we have access to whole-genome variants, including both SNP and structural variations, potentially in the trillions? These scenarios present computational challenges.

Where might we find solutions for these realized potentials? Could machine learning or deep learning offer help? I would like to explore these foreseeable challenges and propose potential solutions by summarizing recent advancements from published research and my own work.

Keywords: genomic prediction, genome-wide association study (gwas), genomic selection, conifer, machine learning

TOPIC 3.
**REPRODUCTIVE PHENOLOGY AND SEED
PRODUCTION**

KEYNOTE PRESENTATION

REPRODUCTIVE PHENOLOGY, GENETIC DIVERSITY AND SEED PRODUCTION IN CLONAL SEED ORCHARDS

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Operational breeding to improve forest trees has been practiced during a century, and large gains are realized in forests worldwide. Most programs were from their initiation challenged by three problems: (i) need to develop multiple breeding and deployment zones in order to accommodate climate driven genotype by environment interaction; (ii) problems with precision of selection for traits expressed at mature stage due to moderate/low juvenile-mature correlation; and (iii) complication of fast and large-scale propagation of the improved trees at low costs. Vegetative propagation techniques have been developed for many species to address the propagation problem, but only employed for a few genera and selected breeding zones - mainly due to the higher cost compared to seedling production. Instead, seed orchards have since the initiation of the breeding work been the most important vehicle for deployment of improved germplasm for the majority of species.

The success of the seed orchard also relates to the two first mentioned problems: a consequence of GxE and imperfect selection precision is that tree breeding (with few exceptions) can rarely be practiced based on precision deployment of highly breed varieties. Such genetic precision deployment become even less relevant for long rotation tree species in the face of the partly unpredictable future climate change. Instead, resistance and resilience facilitated by genetic diversity can become a breeding objective per se, and here the deployment through seed can be an advantage.

However, deployment of seed orchards have not been without problems. Generative reproduction among the trees in the seed orchard is by nature a random process, and the outcome in terms of seed crop varies according to a number of factors that partly depend on biology of the species, but also influenced by the location, genetic composition, design, growth conditions, surrounding forests and yearly climate fluctuations in the seed orchard. Factors that can lead to major deviation of the seed crop from the expected genetic composition and genetic worth, and therefore are collective termed as seed orchard dysfunctions. Here, we review examples where variation in the reproductive phenology have led to such dysfunctions. Examples include problems with low hybrid percent in seed orchards, high selfing rates, increased pollen contamination, but also less visible effects on genetic diversity and genetic worth. Besides addressing the implications of these non-random mating effects, we open for a discussion on how to address the issues in present and future breeding activities.

Keywords: reproductive phenology; non-random mating; genetic diversity; seed orchard dysfunctions

TRANSCRIPTOME AND MORPHO-PHYSIOLOGICAL ANALYSES REVEAL FACTORS REGULATING CONE BUD DIFFERENTIATION IN QINGHAI SPRUCE (*PICEA CRASSIFOLIA* KOM.)

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Coniferous species are mostly monoecious. Their usage in forest plantations is largely limited by the low and unstable yields of seed cones. Gibberellins (GAs) and ethylene are central gender regulators in flowering, although knowledge about their metabolism is limited in conifers. Here, we explore gender determination regulators, especially GA and ethylene metabolism genes, in Qinghai spruce (*Picea crassifolia* Kom.) by transcriptome analysis of female, male and vegetative buds during bud formation and development. Year-round observations were performed to determine the window of bud differentiation. Samples of buds before and after differentiation were collected for RNA sequencing, which showed that the bud gender determination process occurred before the morphological differentiation of female, male and vegetative buds. Sixteen homologs of GA metabolism genes were identified in the transcriptome. Concentrations of GA1, GA3, GA9, and GA15 were identified and quantified using ultra-high-performance liquid chromatography—high-resolution mass spectrometry (UPLC-HRMS). Furthermore, phylogenetic analysis was performed on 9 putative *ACC synthesis* (*ACS*) genes identified in the transcriptomes, and quantitative real-time PCR was conducted to examine gene expression. Our study provides information on GA and ethylene metabolism genes in female, male and vegetative buds during bud differentiation. Overall, fine-tuned regulation of GA metabolism contributes to both the reproductive transition and gender differentiation in this species, whereas ethylene might affect male cone bud formation. The current findings could further our knowledge about the regulatory mechanisms underlying the gender differentiation of cone buds.

Keywords: conifer, ethylene, gibberellin metabolism, RNA-seq, gender differentiation, qinghai spruce

PHENOTYPIC PLASTICITY OF SELECTED REPRODUCTIVE TRAITS OF EUROPEAN LARCH CLONES ON SEED ORCHARDS

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European larch in Poland naturally forms stands mainly in the mountains, in the rest of the area it usually occurs as an admixture in the forest stand. Phenotypic plasticity of selected reproductive traits of European larch clones was studied at seed orchards in Sekocin Stary and Oleszyce. The aim of the study was to determine the genetic effect and the influence of phenotypic plasticity on traits related to flowering and yielding of European larch. Evaluation of variation in flower development rate was performed once in the middle of the flowering period in spring 2020, 2021 and 2022. Both female and male flowers were counted, classifying the intensity of flowering in individual trees. The averaged flowering onset date was determined based on remote observation with cameras. Flowering date data was also given as the number of days since the beginning of the year, the so-called "DOY". The values of air temperature and the intensity of light reaching the plants were recorded with sensors, as well as the date and amount of precipitation. During the growing season, cone development and graft growth were monitored. In the plot in Oleszyce, weak flowering was observed, In the plot in Sekocin, abundant flowering occurred. In Sekocin, cones were harvested from each tree separately, and they were mechanically shelled in the IBL laboratory. Flowering during the analyzed period was variable. Yielding was also variable, with the three clones yielding the most abundantly yielding from 23% to 71% depending on the year. The number of flowers did not determine the number of cones maturing in autumn. The yield was also affected by environmental conditions, especially low temperatures during the flowering period and pests damaging the cones. Fertility varied from year to year and modified the value of the effective population size index calculated with the flowering and cone production taken into account. In order not to narrow the genetic variability of forest reproductive material, cones should be collected also from those clones that produce few cones. The study was conducted on a relatively young seed orchard, six years old in 2022. Thus, it can be assumed that with the development of trees in the following years, the share of clones in cone production will change.

Keywords: european larch, seed orchards, flowering and cone production, phenotypic plasticity

TOPIC 4.
MATING PATTERNS & GENE FLOW
USING MOLECULAR MARKERS

KEYNOTE PRESENTATION

UNDERSTANDING MATING DYNAMICS AND MITIGATING POLLEN CONTAMINATION IN FOREST TREE SEED ORCHARDS: INSIGHTS AND STRATEGIES

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Forest tree seed orchards are artificial populations of genetically superior individuals that play a crucial role in the production of high-quality seeds for reforestation and afforestation programs worldwide. In the pre-genetic-marker era, seed orchards were assumed to act as closed, panmictic populations with equal reproductive success among parents and with no gene flow from external pollen sources. Meeting these assumptions would ensure that the genetic gain attained by breeding would be efficiently transmitted to the next generation, i.e., into seed orchard crops. Many studies published to date have shown that parental reproductive success may be highly variable and that gene flow from undesired pollen sources, a.k.a. pollen contamination, can be substantial. Since the realized genetic gain can be considerably reduced, it is important to monitor mating patterns in seed orchards and thereby control the genetic quality (gain and diversity) of their crops.

With the development of genetic markers, the theoretical assumptions as well as the efficiency of measures proposed to enhance desired crosses and reduce pollen contamination in seed orchards could be verified. First attempts to unravel mating patterns and quantify pollen contamination in seed orchards date back to the late 1970s when allozyme markers were introduced. Allozymes remained in use for over two decades, but due to their low resolution, they were gradually replaced with much more powerful microsatellites (SSRs), which, along with the rapid evolution of various statistical approaches, were capable of providing a much more detailed picture of seed orchards' mating dynamics through pedigree reconstruction. Recently, SNP arrays that have been (and are being) developed for a number of commercially important forest tree species make it possible to affordably and rapidly screen seed orchard seed lots and evaluate the orchards' genetic efficiency.

Keywords: mating pattern, gene flow, seed crop, genetic quality, genetic marker

LOW INFLUENCE OF EXTERNAL POLLEN SOURCES IN SWEDISH NORWAY SPRUCE SEED ORCHARDS

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Seed orchards are currently the most efficient tool to transfer genetic gain from tree breeding efforts into production forestry. One concern has been the gene flow of unimproved pollen into these orchards that dilutes the expected gain. To estimate this inflow of background pollen we reconstructed the pedigree of three crops from each of three different Norway spruce seed orchards and four Scots pine seed orchards in Sweden. We utilized genotyping-by-sequencing to achieve high confidence estimates of pedigrees, based on more than 6000 SNPs, among seedling from these crops. Although background pollination appears to be a much smaller and more stable in Norway spruce than in Scots pine orchards, we see a variation in genotype representation of the orchard clones in individual crops. The variation of fecundity across years for individual genotypes is smaller than between genotypes. This suggests that crops from an orchard can have slightly different properties over time and evaluating fecundity among orchard genotypes could help adjust ramet numbers to produce the expected genetic composition of a crop.

Keywords: *Pinus sylvestris*, *Picea abies*, crop contribution, fecundity, background pollen, genetic composition, genetic gain

ENSURING RELIABILITY AND QUALITY OF NEW SEED ORCHARDS ALONG THE TRACK FROM THE BREEDING POPULATION TO SEEDLOTS IN FRENCH VARIETIES OF LOBLOLLY PINE AND DOUGLAS FIR

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Beside cooperative breeding of *Pinus pinaster* (5th generation in preparation), FCBA has designed and installed several Loblolly pine seed orchards enabling to supply seeds to the forest owners. Moreover, a new French Douglas fir breeding population has been designed and is being archived and propagated for testing in the frame of a cooperative breeding initiative developed by a public-private consortium. To help the proper deployment and monitoring of the development of these seed orchards, we initiated a project aiming to define new kind of cheapest and powerful molecular markers for several aspects of design and deployment of forest tree varieties. While the final aim will be the deployment of a mid-size multiplex (a few hundred markers) to be genotyped by regular HT sequencing, we started to select candidate SNPs from ddRADseq and Axiom genotyping in two broadleaves hybrid systems as well as in two conifers. In Douglas fir, a partnership with the OSU-PNWTIRC allowed us to identify good candidates SNPs from the Axiom genotyping data of hundreds of full-sib families. Then several iterations of multiplex design and genotyping in the Sequenom mass-array enabled us to check the plant material established at two field repositories of the brand-new French breeding population of this species. A very few identification errors were detected. Very good genotyping success was also recorded from southern US provenances although these were not at all represented in the reference data. In the Loblolly pine (*Pinus taeda*) case, we got access to data from the NCSU-Tip. The design and validation of SNP-multiplexes was done with plant material of three new seed orchards established by FCBA on its own breeding program for several industrial consortia, checking plant identity at all stages from clonal repositories to freshly grafted material and various series of planted sector of same orchards.

Keywords: SNP, pollen flow, grafting, monitoring, breeding population

THE CONTRIBUTION OF INDIVIDUAL POLLEN DONORS IN A HYBRID LARCH ORCHARD FROM NORTHERN GERMANY

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Offspring from matings between European larch (*Larix decidua* Mill.) and Japanese larch (*Larix kaempferii* Lamb.) is called 'hybrid larch'. These hybrids typically show vigorous growth due to the heterosis effect, combining both parental species' suitability to different environments and resistance characteristics. In progeny tests hybrids achieved 120-180 % of basal area increment relative to pure species. In Germany, hybrid larch seeds are produced in seed orchards. Here, we analyse a seed orchard composed of one European larch replicated 500 times acting as mother clone and at approval in 1992 still 195 replicated Japanese larch clones from 36 stands in the natural range as pollen donors. At time of approval hybrid proportions were reported as 82-85 % according to isoenzyme analysis. In later years this proportions decreased to 41 % in 2016. Due to land use change starting 2010 and salvage cuttings in response to calamity the orchard got successively reduced from 1.7 to 0.8 hectares with 85 remaining Japanese larch clones in 2021, which we sampled for genotyping.

Here we use 13 microsatellites to reconstruct the pedigree of three reproduction events, 2009, 2013 and 2018 (sample sizes of 160 - 200 seedlings), in order to identify the contributions of individual fathers, rate of selfing and impact of size reduction. For 2009, before size reduction, we could identify 37 Japanese larch clones as pollen donors. At presentation results of the three progenies will be compared.

To identify the overlap in flowering period between parental clones, we further provide observations of spring flushing and flowering on grafts produced from the remaining orchard parents, as well as from progenies of the original Japanese stands.

The outcomes of this study will be used to establish optimised, highly productive seed orchards, where a European larch will be combined with the most fertile pollen donors.

Keywords: bud flush, grafting, *Larix x europelis*, parental analysis

TOPIC 5.
**GENETIC GAIN & DIVERSITY OF SEED
ORCHARDS CROPS**

KEYNOTE PRESENTATION

ESTIMATION OF GENETIC GAIN AND GENE DIVERSITY FOR SEED ORCHARD CROPS

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Seed orchards are the major tool for deploying the improvement generated by tree breeding programs and assuring the consistent supply of genetically improved seed. Attainment of genetic gain and monitoring of gene diversity through selection and breeding were studied considering the factors: selection intensity; genetic value; coancestry; fertility variation; and pollen contamination. The optimum goal of a seed orchard is achieved when the orchard population is under an idealized situation, i.e., panmixis, equal gamete contributions from all parental genotypes, non-relatedness and no pollen contamination. In practice, however, due to relatedness among parents, variation in clonal fertility and ramet number, and gene migration from outside, the realized genetic gain and gene diversity deviate from the expectation. In the present study, the genetic value of seed orchard crops (genetic gain) could be increased by selective harvest, genetic thinning and/or both. Status number was used to monitor the loss of gene diversity in the process of forest tree domestication, and calculated to be reasonably high in most seed orchards. Fertility of parents was estimated based on the assessment of flowering or seed production, which was shown to be under strong genetic control. Variation in fertility among orchard parents was a general feature and reduced the predicted gene diversity of the orchard crop. Fertility variation among parents could be described by the sibling coefficient. In calculating the sibling coefficient, it was possible to consider, besides fertility variation, the phenotypic correlation between maternal and paternal fertilities, and pollen contamination. Status number was increased by controlling parental fertility, e.g., equal seed harvest, mixing seed in equal proportions and balancing parental contribution. By equalizing female fertility among over-represented parents, it was possible to effect a favorable tradeoff between gene diversity and seed production. If the status number of orchard crop is not large enough, loss of gene diversity, random drift in gene frequency and potential inbreeding problems could occur in subsequent generations. Genetic loss or erosion did not seem to be alarming during the domestication of forest trees, because a large number of parents are commonly used in first-generation seed orchards. An understanding of reproductive processes and monitoring of the impacts of the management practices are essential to maximize genetic gain and to maintain sustainable gene diversity in seed orchard programs.

Keywords: status number, effective population size, group coancestry, inbreeding, fertility variation, ramet variation, gene migration, sibling coefficient

EVALUATION OF GENETIC DIVERSITY AND GAIN IN *EUCALYPTUS CAMALDULENSIS* SEED ORCHARDS OF TWO GENERATIONS

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Genetic diversity and genetic gain were evaluated in seed orchards of two generations in a breeding programme of *Eucalyptus camaldulensis* in India. Genetic gain trials compared the performance of four first-generation (F_1) seedling orchards (seed production areas: F_1 SPAs 1-4) with the control, a bulked natural-provenance seedlot. Fertility was low in three orchards (F_1 SPAs 1,3&4) with less than 30% flowering trees at five years, whereas 73% trees flowered in F_1 SPA2, with an outcrossing rate of 86%. Fertility variation was higher in the three low flowering F_1 SPAs (sibling coefficient, $\Psi = 5-11$) than the high fertility F_1 SPA2 ($\Psi = 2.27$). At three years, seed crops of the four F_1 SPAs gave no significant gain over the natural provenance seedlot, with survival ranging from 79-93%.

Five second-generation (F_2) orchards were developed: (a) two F_2 SPAs (1&2): by thinning two F_1 genetic gain trials (b) F_2 SPA3: by thinning one seedlot of the high flowering F_1 SPA2, and (c) two F_2 CSOs (1&2): by converting two clone trials of F_1 selections. Fertility variation was high in the F_2 SPAs also (Ψ , 9-14) with 26% fertile trees, but fecundity was greater than the natural provenances. Effective population size of F_2 SPAs (1&2) was higher (N_s , 95 and 74) than the F_2 SPA3 ($N_s = 39$). The F_2 CSOs had 81% fertile trees but fertility was highly skewed in CSO2, with a lower effective population size ($N_s = 2$) than CSO1 ($N_s = 11$). The progeny of the two F_2 SPAs -1 &2 had the same genetic diversity (H_e - estimated using SSR markers), but better growth than the natural provenance at 3 years. CSO2 had the lowest H_e and poor survival at three locations. Genetic composition and fertility status of the orchard trees affected the performance and genetic diversity of the progeny.

Keywords: seed production area, clonal seed orchard, fertility, provenance, survival

HOW DO PHENOTYPES OF SEED ORCHARD (IMPROVED) VS WILD (UNIMPROVED) SEEDLINGS OF PINUS CONTORTA (LOGEPOLE PINE) DIFFER UNDER COMMERCIAL GREENHOUSE CONDITION?

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Reforestation in Alberta (AB) relies on greenhouse grown seedlings vs direct seeding. Current policies recommend employing improved (seed orchard) seedlots for reforestation whenever feasible to provide greater wood volume on the land-base. The selection trait for volume is height, which can be challenging as nursery managers strive to achieve uniformity among both improved and unimproved (wild collected) seedlots based on the same criteria or 'specs' for retaining seedlings at lifting (height: 7-23cm, root collar diameter (RCD) > 2.2mm).

We assessed performance in both seedlot types of lodgepole pine from germination to lifting (~6 months). Seedlings from eight seedlots (four improved and four unimproved) were monitored and a subset of seedlings were harvested at lifting. During lifting, seedlings that did not meet specs were culled and discarded. All discarded seedlings and 30 specs seedlings were collected for further measurement. A minimum of 100 culled and the 30 specs seedlings, per seedlot, were measured for height, RCD, above- and belowground biomass, and leaf area. We compared height and RCD performance between: 1) improved vs unimproved seedlot seedlings; and 2) culled vs specs seedlings within seedlots. Using Welch's t-test and ANOVA, we showed that seedlings from all four improved seedlots were taller than unimproved seedlings ($p < 0.001$), culled of two out of four improved seedlots were significantly taller with larger RCD than their corresponding specs seedlings ($p < 0.001$), and three out of four and two out of four culled seedlings of unimproved were taller with larger RCD respectively than their specs seedlings ($p < 0.001$).

Root-to-shoot (R:S) ratio showed three out of four improved specs had higher root biomass than the culled whereas two specs of unimproved had a higher ratio than the corresponding culled ($p < 0.05$). Overall biomass results showed that most of the improved (both culled and specs) seedlots had a significantly higher R: S ratio vs the unimproved seedlots.

Keywords: tree improvement, improved & unimproved seedlots, nursery production

VARIATION OF STROBILI AND CONE PRODUCTIONS IN CLONAL SEED ORCHARDS OF ANATOLIAN BLACK PINE

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Anatolian black pine [*Pinus nigra* Arnold subsp. *pallasiana* (Lamb.) Holmboe] is an economically and ecologically forest tree species by 4.2 million ha natural distribution which of 31.9% is unproductive. The species is used widely in afforestation practices and conversion of unproductive forest to productive forest by genetically improved seed crop produced from seed orchards or other seed sources. Numbers of cone, female and male strobili were studied in three seed orchard populations consisting of 30, 30 and 34 clones, and five grafts of each clone and each seed orchard for two consecutive years (2022-2023). Variation, broad-sense heritability (H^2) (clonal repeatability) and correlations at graft and clone levels among cone and strobili productions were estimated in the present study. Strobili and cone productions were more changeable. They varied among orchards, among clones within orchard and between years within orchard. Significant ($p < 0.05$) differences were found for most characters among clones within orchard, and between years. Relations among the characters changed for the characters, year and orchard. However, female strobili seemed a better predictor for cone production. Variations among grafts were higher among clones for all characters in seed orchards and years. The heritability in broad sense was on average below 0.5 for all characters in individual seed orchard.

Results of the study were discussed based on future practices of seed orchards.

Keywords: correlation, genetic, graft, heritability

ADVANCED - GENERATION SEED ORCHARDS OF EUROPEAN SILVER FIR IN ROMANIA – ACHIEVEMENTS AND PERSPECTIVE

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The European silver fir (*Abies alba* Mill.) is one of the most important conifer tree species in Europe. In Romania, it is a main component of mountain forests and has manifold ecological, economical and soil protective functions. Environment changes will pose severe pressures for European silver fir populations and therefore, producing of the improved forest reproductive material, that has high growth and adaptive capacity, for regeneration and afforestation program is required. In Romania, there are 92.1 ha of the first-generation seed orchards of European silver fir which comprise of vegetative copies of plus trees selected in natural populations. The seed orchards are 40 years old and therefore, the establishing of the second generation of seed orchards is necessary. To address these problems, since 2007 an advanced-generation breeding plan for European silver fir has been started. To evaluate the breeding value of the plus trees from the first-generation seed orchards, eight progeny tests based on both controlled and open pollinations have been established. The objectives of this study were: to investigate the genetic variation of the quantitative and adaptive traits in five full-sib and three half-sib progeny tests of European silver fir, to determine and to compare the genetic parameters from control-pollinated and open-pollinated progenies, to assess the genetic correlations among traits, to evaluate genotype x environment interaction, to determine the genetic gain under different breeding strategies and to select the candidate genotypes for the advance-generation seed orchards. To ensure the maximum genetic gain and gene diversity in the second-generation seed orchards crops, the marker-assisted selection is applied at many stages of the breeding program, such as identification of genotypes, pedigree reconstruction and selection of the superior parents. Results will have extreme importance in the species' breeding program. This information will complete the first breeding generation of European silver fir in Romania and will be necessary to advance to the second breeding generation.

Keywords: european silver fir, genetic parameters, genetic gain, adaptive capacity, multi-trait selection

A LOW INPUT - HIGH SELECTION INTENSITY - APPROACH FOR RAPID DEVELOPMENT OF ASH TREES WITH HIGH TOLERANCE TOWARDS ASH DIEBACK BASED ON BREEDING SEED ORCHARDS (BSOS)

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Ash dieback (ADB) caused by *Hymenoscyphus fraxineus* has become a major problem across Europe. Young ash trees often die within few year, and trees of all ages experience severe damage and increased mortality. Fortunately, additive genetic variance in tolerance seem moderate to high with limited GxE interaction, where young trees often reveal their level of tolerance within few years. Combined with early flowering, these features make phenotypic selection suitable for breeding for increased ADB tolerance with seedling seed orchards suitable for commercial seed procurement. We present a low input breeding program for ADB tolerance based on the breeding seed orchard (BSO) concept. The program was initiated 2001 as a joint effort between the Danish Nature Agency and University of Copenhagen. Seed collected after open pollination from 100 selected trees across the Danish landscape, and a first generation BSO established in 2004 with the putative half-sib families. Selection and generation turn-over was initiated after 14 years of testing, and second generation BSO established in 2022. We present the idea behind use of the low input BSO the concept, expected versus realized gains from first generation, current status for the second generation BSO, and lessons learned in terms of options and limitations from the application of the BSO concept in ash breeding. We also discuss how the BSO approach can be expanded and integrated with other national breeding activities on ash in Denmark, and how the work has recently become part of a region effort across Lithuania, Norway, Sweden and Denmark. Finally, we discuss how the low input breeding can benefit from the various ‘omic tools that have been developed for ash during the recent decade. For further perspectives, we discuss the general value of having BSO based programs for a diverse array of so-called minor tree species in face of climate change.

Keywords: ash dieback, breed seed orchards, seedling seed orchards, resistance breeding, phenotypic selection, *Fraxinus excelsior*

ENHANCING GENETIC IMPROVEMENT CLONAL SEED ORCHARDS OF *PINUS DENSIFLORA* THROUGH FERTILITY-BASED CLONE SELECTION

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This study examines the influence of variations in clonal fertility on the genetic progress within a second-generation seed orchard of *Pinus densiflora* located in Chuncheon, Republic of Korea, established in 2014. The research focuses on 42 clones, encompassing 1,300 ramets, from 2020 to 2023, to evaluate reproductive traits for insights into genetic variability and inheritability. Through Analysis of Variance (ANOVA) and comparison of Specific Combining Ability (SCA)-derived genetic gains against those adjusted for fertility traits, genetic variation was noted significantly. Heritability estimates across the years for female and male strobili, conelets, and cones were robust (0.69 ± 0.09 , 0.58 ± 0.13 , 0.63 ± 0.11 , and 0.55 ± 0.15 , respectively). Despite this, fertility-adjusted genetic gains showed a declining trend from 21.02% in 2020 to 10.77% by 2023. In contrast, estimates of gene diversity increased during this period, influenced by the production rates of female and male strobili, conelets, and cones ($0.41 \pm 0.02\%$, $2.40 \pm 2.07\%$, $0.70 \pm 0.52\%$, and $0.86 \pm 0.72\%$, respectively). The findings highlight the necessity of strategic clone management, including reselection based on fertility and genetic gains and the adoption of genetic thinning or selective harvesting, to enhance the quality and quantity of cone production. This approach is pivotal for the development of future orchard generations, especially in selecting clones and designing layouts that consider the terrain's gradient.

Keywords: fecundity variation, *Pinus densiflora*, clonal seed orchard, seed orchard management, genetic variability

GENETIC DIVERSITY OF SILVER FIR AND EUROPEAN LARCH SEED ORCHARDS IN ROMANIA - SUPPORT FOR ADVANCED BREEDING GENERATIONS

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Forest tree breeding is an important component of forestry aiming at increasing the potential for the accumulation of high-quality wood mass, resistance to diseases and pests, as well as high adaptability to drought and late frosts. Microsatellite genetic markers were used for the evaluation of genetic diversity of first-generation seed orchards, fingerprinting the clonal genotypes and evaluating the genetic relationships.

Five silver fir (*Abies alba* Mill) and five European larch (*Larix decidua* Mill) seed orchards have been selected. We successfully genotyped 185 clones of silver fir and 246 European larch clones, respectively. In both species, the provenance mentioned for the clone origins recorded at the establishment of the seed orchards was not always the same with the results based on molecular markers; in this regard, the present study is important as it clarifies this information.

In silver fir, we analyzed seed orchards of different sizes and clonal compositions located in several ecological regions of Romania. The loss of alleles with the decreasing number of the clones in seed orchards and an increased value of genetic diversity when the number of clones is comparable, but the origin is different, was observed. The results revealed significant levels of genetic diversity ($H_E = 0.598$) within the seed orchards, with implications for population structure and long-term sustainability.

A high level of genetic diversity was detected in European larch seed orchard ($H_E = 0.764$). The seed orchards showed heterozygote deficiency, similar to other studies in European larch or other larch spp., presumably associated with a strong selective pressure on populations in a highly fragmented area. The presence of clones of autochthonous origin increased the allelic richness in the seed orchards. The degree of differentiation between individuals within the seed orchards was similar to that of populations originating from the Tyrolean Alps and the Southern Carpathians. The results may facilitate the transition to advanced-generation seed orchards and contribute to enhance conservation efforts.

Keywords: european larch, silver fir, genetic diversity, genetic structure, microsatellite nuclear markers

A NOVEL ALLELIC-BASED STOCHASTIC SIMULATION MODELING FOR SEED ORCHARD DEVELOPMENT

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Seed orchards of forest trees consist of genetically improved genotypes with superior economically important traits. Many countries have ongoing seed orchard development and management programs prioritizing genetic gain while accounting for genetic diversity and minimizing the detrimental inbreeding effects. The genetic quality and diversity of seed crops produced from seed orchards are influenced by various factors, including the genetic superiority of the selected parents and their genetic kinship, the extent to which they contribute gametes to the final seed crop, the degree of gene flow from outside pollen (pollen contamination), and the parental fertility variation. The application of high-throughput phenotyping and SNP marker-based genotyping have improved the accuracy of genomic predictions in seed orchard production. In this study, we construct seed orchards using genomic data of tree species with different-sized parents under the “Breeding without breeding” methodology. We present allelic-based stochastic simulation models that involve three stages: (a) population simulation, (b) genetic evaluation, and (c) seed orchard selection and deployment. We simulate the breeding population and the respective mating scheme in the first stage. Then, the genetic evaluation (done through internal calls to the ASReml software). Finally, we select the top-ranking individuals from the current cycle's candidates based on mathematical optimization. We aim to maximize the genetic gain per unit of the effective population size while considering additional constraints. The variance components of genetic parameters are estimated using phenotypic data of randomly selected progeny sub-populations and their inverted numerator pedigree relationship (pedigree) matrix. The results of the effect of seed orchard sizes and their random progeny tree selection on the estimated and actual narrow-sense heritability using the identity-by-descent and genomic relationship matrices are presented. According to this study, allelic-based simulation models accounting for the complex genetic architecture of quantitative traits are efficient and beneficial for breeders to make decisions regarding the deployment of seed orchards and genetic evaluations.

Keywords: seed orchard, stochastic simulation, heritability, breeding without breeding, genetic gain and diversity

GENETIC PARAMETERS AND GENETIC GAINS FROM HALF-SIB AND FULL-SIB PROGENIES OF *LARIX DECIDUA* (MILL) SEED ORCHARDS IN ROMANIA

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In Romania, European larch (*Larix decidua* Mill) represents an important forest tree species and therefore its genetic improvement program began in the 1960s. Over 500 plus trees were selected in natural and artificial stands. Twenty-six seed orchards (134 ha) were established between 1964 and 1980 and they are currently the main sources for producing genetically improved forest reproductive material in Romania.

In order to complete the first cycle of the larch breeding program, eight progeny trials were established in the period 2011-2019 based on both controlled and open pollinations in first-generation seed orchards. In these trials, 102 half-sib families, 35 full-sib families and 112 clones of European larch are tested.

The objectives of this study were: to investigate genetic variation in growth, branch characters and bud flushing in European larch half-sib and full-sib progenies; to determine and compare the genetic parameters from half-sib and half-diallel tests; to evaluate genotype x environment interaction; to evaluate the genetic correlations and to estimate the genetic gains. Information will be necessary to formulate optimum strategies for this species and to select the genitors for next breeding generation.

Results provide important knowledge concerning genetic parameters of the European larch and are extremely important for both breeding program and conservation of forest genetic resources under climate changes.

Keywords: European larch, genetic parameters, genetic gain, early selection

TOPIC 6.
**EFFECT OF MANAGEMENT ON CONE
PRODUCTION**

KEYNOTE PRESENTATION

MANAGEMENT OF THE FOURTH-CYCLE SEED ORCHARD IN CHINESE FIR

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Chinese fir (*Cunninghamia lanceolata* (Lamb.) Hook.) is the most important timber species in China, accounting for 20% of the plantation area and contributing to 25% of the wood volume in Chinese plantations. We collected 608 elite germplasms from 1st -3rd cycles progeny and provenance trials and grafted them in the 4th cycle clonal archive, in Yangkou Forest Farm in Fujian province. Through the analysis of genetic diversity and structure within the 4th cycle Breeding Population of Chinese fir, utilizing 322 germplasms, we discovered that a set of 50 representative individuals could effectively balance genetic gain with similar genetic diversity. In our investigation into the selection and deployment of parents for the 4th generation dwarf seed orchards of Chinese fir, we observed that after dwarfing, some of the finest candidate germplasms were unable to produce cones in the lower portion of the crown. Consequently, we selected the top 28 clones exhibiting robust cone setting at an early age after dwarfing for inclusion in the fourth-circle seed orchard. Finally, we have established two seed orchards comprising 48 selected germplasms from the 4th archive at Yangkou and Guanzhuang Forest Farm in Fujian province.

Furthermore, we conducted an analysis of flowering phenology and synchronization indexes of Chinese fir within our clonal archive. The flowering synchronization index of 58% clones was higher than 0.5. The early type and middle type possess higher flowering synchronization indexes. Utilizing aerial drones and vacuums to aid in pollen pollination in a Seed orchard, we succeeded in enhancing seed production and quality by utilizing elite pollen from the best elite trees in other seed orchards.

Keywords: *Cunninghamia lanceolata*, fourth-cycle dwarf seed orchard, flowering phenology, selection, deployment

HIGH STAND DENSITY IMPROVES SEED PRODUCTION IN SEED ORCHARDS OF THE MASTING SPECIES *PICEA ABIES*

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Reproduction in masting species is characterised by long intervals between good cone and seed production years, and only sparse reproduction between mast years. Examples of economic important tree species besides *Picea abies* is *Quercus robur*, *Fagus sylvatica*, *Picea glauca*, *Pinus nigra* and *Castanea sativa*. The physiological mechanisms behind masting, and how these are linked to internal resource status and external weather factors, is still a subject of scientific exploration and debate, as is the effect of climate change on masting. This study investigates cone production in one operational seed orchard in Sweden which was established with two different spacings and has since been subject to three tree thinning experiments. The spacings before thinning varied between 800 and 400 stems · ha⁻¹, and then thinning reduced the stand density in all trials to half, i.e. between 400 and 200 stems · ha⁻¹. In all three experiments cone production per tree was equal in un-thinned and thinned treatments, both in mast years and in non-mast years. Thus, the cone production per unit area was twice as high in the un-thinned areas. The conclusion from these experiments is that the establishment of *Picea abies* orchards with wide tree spacing is both a misuse of good orchard locations and bad economics.

Keywords: norway spruce, cone production, thinning

CONIFER SEED ORCHARDS IN THE US PACIFIC NORTHWEST

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Seed orchard experience in the US Pacific Northwest goes back to the early 1950s. Early grafted Douglas-fir orchards ran up against delayed graft incompatibility, which led to a shift to establishing full-sib seedling seed orchards in the 1970s until graft compatible rootstock were developed.

Graft incompatibility has not been an issue for other conifer species. Seed orchards were established in a variety of locations, including close to the Pacific Ocean and deep in the forests and at high elevations. The best results are obtained in mild agricultural locations with spring drought, such as the Willamette Valley in Oregon, and comparable environments in western Washington. Grafting on rootstock established in the field was common at one point, but current grafting is mostly in greenhouses on potted rootstock. Grafted ramets are usually held another 8 to 12 months before out planting. Well-managed “pot grafting” or “bench-grafting” programs can lead to over 95% graft survival, rapid growth in the first year, uniform orchards, and greatly improved working conditions for grafters compared to field grafting. Orchard spacing varies from as low as 120 ramets per hectare (rph), to 480 rph to make better use of growing space in the first decade. Ramets established with mature phenotypes from high elevations have been established as tight as 652 rph. One entity is focusing on hedged orchards at even higher densities. Tools (such as double-overlapping girdling) to produce consistent cone crops and high seed yields (insect control) have been developed. Orchards are also successfully established and managed for western hemlock, noble fir, western red cedar, sugar pine and ponderosa pine. As average temperatures go up, extreme heat events occur, and summer droughts intensify due to climate change, some orchard locations may become less viable; dry-site orchards dependent on irrigation may face issues as well.

Keywords: Pacific Northwest conifers, grafted seed orchards, cone production, stimulation

SITE VARIATION IN CONING AND NUTRIENT CONTENT IN DANISH NORDMANN FIR SEED ORCHARDS

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In Denmark, breeding for improved Christmas tree quality in *Abies nordmanniana* was initiated in 1992. Today, more than 60 hectares of seed orchards have been established. Generally, seed production are abundant, and up to several hundred kilos of seed per hectare - meaning several tons of cone material - are removed on a yearly basis. Thereby, causing concern of the long-term sustainability in nutrient supply for the seed producing grafted trees and its consequences for seed production. The purpose of the study was to quantify site differences in cone setting, estimate nutrient content in cones, dry matter content and finally estimate amount of nutrients removed by the cone crop. Five grafted seed orchards were included in the study, and the scion material originates from two Danish approved stands F.526 and F.527 Tversted dating back to 1902. All orchards are producing seed in commercial quantities. Individual tree cone counting were carried out in the five orchards in year 2022 and 2023. Nutrient analyses, measurements of cone weight and dry matter content based on three cone samples per site, and two samples of tree needles and soil were carried out in 2022 (all samples pooled values of 20 random trees). Cone nutrient content across sites was remarkably uniform despite soil differences and geographic distance. Nutrient content of needles varied more across sites than for cones, especially for manganese (Mn) and iron (Fe). Comparing the nutrient content as the rate between cones and needles the most striking is a substantial surplus of potassium (K) in the cones. Cone counts per tree ranged from 29 to 45 and 37 to 62 in years 2022 and 2023, respectively. Average cone weight across sites ranged in 2022 from 154 to 177 grams per cone. Implications for seed orchard management and fertilization will be discussed.

Keywords: fertilizing, nutrient content, strobili

MASS-PRODUCTION OF HYBRID LARCH FOREST REPRODUCTIVE MATERIAL: IS ADVANCED-GENERATION HYBRIDISATION SEED ORCHARDS AN OPTION?

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Highly performant hybrid varieties of European x Japanese larches have been selected across Western Europe. Their mass-production has been so far quite exclusively achieved through first-generation hybridisation seed orchards, with some attempts through vegetative propagation. Whereas technically feasible either through cuttings (bulk propagation) or somatic embryogenesis, vegetative propagation remains indeed so far commercially unsuccessful. Mass-production through seed orchards remains thus the only commercial option. Several first-generation seed orchards have been established across Europe combining various sets of clonal parents and planting designs. Among other things, their success is nevertheless mitigated by the rate of hybrids in seed lots. An alternative is to proceed through composite breeding and advanced-generation hybridisation seed orchards. First such seed orchards have been planted in Belgium and in France. Preliminary results from a first-/second-generation hybrid factorial design will be presented which shed light on the potential of such seed orchards.

Keywords: Larix, hybridization, seed orchard, composite breeding, hybrid depression

TOPIC 7.
**RELATIONSHIP OF THE SEED ORCHARDS
WITH OTHER FOREST ISSUES**

KEYNOTE PRESENTATION

UNVEILING THE SPECTRUM: ANALYZING GENETIC PARAMETERS OF HYPERSPECTRAL REFLECTANCE AND LEAF FUNCTIONAL TRAITS IN REPLICATED CLONAL SEED ORCHARDS WITH ANNUAL DYNAMICS

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High-throughput phenotyping (HTP) revolutionizes the assessment of organisms on a large scale, employing advanced technologies and data analysis. In forestry, it plays a pivotal role in studying trees, utilizing cutting-edge imaging, sensing, and robotic technologies. By intensifying data collection, particularly in forest genetics and tree breeding, HTP enables the identification of patterns and correlations more efficiently than traditional methods of manual measurements, which are laborious, expensive, and prone to errors. The rising costs of phenotyping hinder the full utilization of advancements in molecular and genomic analyses. A specific example from Scots pine and Norway spruce seed orchards in the Czech Republic illustrates that HTP uncovers broad-sense heritability in spectral reflectance and genetic correlations among spectra and ground truth measurements. The whole spectral range was explored through partial least squares regression (PLSR) to predict photosynthetic pigments content, water content, and needle morphology. The stability of these predictions was further assessed through replicated clonal seed orchards measured in several seasons. The hyperspectral reflectance measurements on leaf (spectroradiometer with a contact probe) and canopy levels (drone-based) revealed valuable information about leaf functional traits (LFT) and, surprisingly, spring phenology, aiding in selecting clones with desired adaptive potential for local conditions. PLSR enabled the prediction of LFT with high reliability (up to 70%), showcasing the power of hyperspectral phenotyping in uncovering LFT variation. Significant genetic correlations of pre-selected (PLSR) wavelengths with LFT indicated potential for indirect selection. These results underscore the utility of leaf traits and leaf-level/canopy reflectance spectra in distinguishing intrapopulation variability. Novel approaches combining reflectance factors and machine learning algorithms provide promising tools for efficient breeding programs. In conclusion, HTP represents a powerful tool for understanding biological systems and exploring genotype-environment interaction. Despite challenges in traditional methods, advancements in technology and data analysis pave the way for innovative phenotyping approaches.

Keywords: seed orchards, spectral reflectance, heritability, genetic correlations, UAV, needle functional traits

UNVEILING NORDMANN FIR SUSCEPTIBILITY TO SILVER FIR WOOLLY ADELGID: INSIGHTS FROM CLONE SEED ORCHARDS

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Nordmann fir is Europe's primary Christmas tree species. Since 1992, Denmark has initiated a breeding program to select high-quality trees, resulting in more than 60 hectares of seed orchards with Danish bred Nordmann fir material. The silver fir woolly adelgid (SFWA) (*Adelges (Dreyfusia) nordmanniana*) poses a significant threat to Nordmann fir Christmas trees, thus resistance to SFWA has become a key selection criterion in the breeding program. Further understanding of tree species' susceptibility to pests and diseases is crucial for effective forest management and breeding. This study evaluates Nordmann fir susceptibility to SFWA by assessing variations in natural infestation levels across three Danish-approved clone seed orchards (CSOs). Evaluation involved 3544 grafted trees from 83 Nordmann fir clones, graded on a scale from 0 (healthy) to 3 for SFWA infestation intensity. Additionally, 1288 three-year-old half-sib seedlings from 40 distinct clones underwent artificial SFWA infestation, monitored over two years and a decade post-infestation for SFWA counts and associated symptoms. Results showed significant clonal variation in SFWA susceptibility, with two CSOs experiencing over 10% natural infestation. A genetic correlation of 0.37 between sites indicated varying susceptibility across environments. Broad-sense heritability for SFWA susceptibility stood at 0.21 (SE: 0.04). Progeny stand analysis revealed substantial genetic control over traits such as SFWA counts, needle curling, and needle discoloration, with significant variations among families. The discoloration score at the tenth-year post-infestation exhibited a high correlation with SFWA counts in the initial two years, suggesting early counting's reliability in assessing susceptibility. Furthermore, high correlations (average 0.77) between CSOs' clone values and parent clone breeding values from progeny trials underscored CSOs' reliability in studying SFWA susceptibility among families. In summary, these findings shed light on the genetic basis of SFWA susceptibility and emphasize the potential of CSOs as tool for identifying SFWA-resistant breeding material.

Keywords: clonal seed orchards, silver fir woolly adelgid, pest problem, resistance breeding

SCOTS PINE SEED ORCHARD EXPERIMENT GIVES INDICATIONS OF POLLEN-MEDIATED TRANSGENERATIONAL EPIGENETIC INHERITANCE

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Forests are amid climate change. The adaptation of a tree individual to a changing climate may be facilitated by the epigenetic events of the genome. Epigenetics is defined as follows: A change in the off-on state of genes that is inherited during cell divisions and is caused by a change in the chromatin structure but is not caused by a change in the DNA sequence. The ability of the Scots pine seedling to survive in a warming climate could be aided not only by its own epigenetic events, but also by the epigenetic information inherited through pollen. Scots pine seed orchards in Finland and Ukraine offered an opportunity to study pollen-mediated epigenetic inheritance. Clonal seed orchards of Scots pine have been established in various locations in Finland in the past decades. In these seed orchards, surrounding pollination may reduce the genetic quality of the seeds. To avoid the effect of surrounding pollination, in 1992 seed orchards of grafted plus trees of Finnish Scots pine were also established in the Vinnytsia region in central Ukraine. The Vinnytsia region is the southern boundary of the natural area of Scots pine and more than 1600 km south of the natural area of Finnish Scots pine plus trees. Similar clonal seed orchards that were established in Ukraine were also established in Finland. Finnish Scots pine plus tree clones that grew for decades in both Ukraine and Finland created an excellent framework for studying the inheritance of environmentally induced epigenetic changes. In this study, certain mother clones of Scots pine were pollinated with both Finnish and Ukrainian pollen of certain father clones, both in Finland and Ukraine. The autumn frost hardening of first-year seedlings grown from the seeds of these controlled crosses was evaluated. Results of freezing tests indicated pollen-mediated epigenetic inheritance.

Keywords: Scots pine, seed orchards, epigenetic, pollen, inheritance

POSTERS

SPRING PHENOLOGY AND GENETIC DIVERSITY OF *QUERCUS ROBURL.* (PEDUNCULATE OAK) IN MOVILENI SEED ORCHARD

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Movileni seed orchard was installed in March 2013 according to a programme which developed a new seed orchards network, and this consists of 2 different national provenances of *Quercus robur* L. (pedunculate oak) with valuable clones which can ensure a large genetic diversity. The aim of the study was to evaluate the genetic variation of pedunculate oak species in the current context of ensuring forest genetic resources, especially for quality wood production. The two provenances (B - Mădârjești, Tecuci Forest District (local provenance) and F - Cobadin, Murfatlar Forest District) were tested according to the spring phenology process from 2016 to 2022, and all data were evaluated to conclude which provenance was better local adapted. Survival rate was lower in B provenance, compared with F provenance (70% and 82%) and in November 2013 (8 months after the establishment) about 23% of planted seedlings were registered dead (7% from F - Cobadin provenance seedlings and 16% from B -Mădârjești provenance seedlings). For B - Mădârjești provenance, the most precocious individuals were registered (DOY 95 was the first date of leafing), and abundant fructification presence was reported (B166 clone). The date of bud burst in spring can vary by as much as 21 days (the earliest date for B provenance was DOY 95, and for F provenance was DOY 115), but the period between earliest and latest individuals can be considered constant (18 days). The differences between individuals' phenology process around this period (2016-2022) were influenced by climate changes and temperature increases, which represent an important aspect of genetic diversity and variation, especially for genetic progress, amelioration and future production of valuable forest genetic resources.

Keywords: spring phenology, provenance, fructification, genetic diversity, production

FORSEE: SEEDS FOR CLIMATE-ADAPTED FORESTS: NEW STRATEGIES FOR MANAGEMENT OF SEED ORCHARDS, SEED STORAGE AND GERMINATION TESTING

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Climate change has serious impacts on Austrian forests: not only are the climatic optima and distribution areas of tree species shifting, but extreme climatic events are increasing, leading to destabilization of forests. Reforestation of disturbed areas thus will become one of the key challenges for forestry in Central Europe in the coming decades. To enable "assisted gene flow", artificial regeneration will become even more important in the future. However, forest nurseries often cannot meet this high demand due to lack of suitable seeds. This is especially the case for oaks and silver firs, whose seeds cannot be stored in the long term, but which are at the same time particularly important for the establishment of climate-resilient forests. Furthermore, in recent years in Austria, decreasing germination percentages and/or harvest failure have been increasingly observed in these species, for which seed demand has steadily increased in recent years. Therefore, it is essential to better understand the physiology of seed production in oak and fir to develop improved management tools to enhance seed production at least in seed orchards. The goal of the project FORSEE is to develop practical concepts for intensifying orchard management to ensure a long-term supply of high-quality forest reproductive material. Results from manipulation experiments in fir seed orchards will be used to develop treatment procedures regarding fertilization and irrigation as needed for continuous yields. The potential and effects of phytohormone treatments on uniform seed yields in oak plantations will be evaluated. In addition, we present the results for a series of fir and oak seed treatment experiments with the goal to optimize harvest and storage logistics for fir and oak.

Keywords: management of seed orchards, seed storage, germination testing, fertilization

DEVELOPMENT OF A NON-DESTRUCTIVE METHOD FOR ASSESSING SPECIES PURITY IN EURO-MEDITERRANEAN FIRS

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Plantations of Mediterranean firs are increasing in Europe because they are more resistant to drought than the native silver fir. However, all the Euro-Mediterranean species (8 species in total) have the capacity to hybridize with each other, which can create difficulties in guaranteeing the species. A simple, rapid and economical method is proposed to evaluate the hybridization rate among seeds intended for nurseries. Genotyping will confirm the predicted hybridization. This study done for the European project “Optforests” about the harnessing forest resources for the forest of tomorrow.

Keywords: seed, pinus, hybridization, spectrometry

ADVANCING SEED ORCHARD DESIGN THROUGH INTEGRATED ARRANGEMENT AND EVALUATION MODELS

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Mitigating the effects of inbreeding is a significant challenge in the management of seed orchards due to its detrimental impact on seed yield and genetic diversity. This study presents an integrated approach for seed orchard arrangement and evaluation to facilitate decision-making in orchard design. The arrangement process involved the utilization of existing models such as the Optimum Neighborhood Algorithm (ONA), FG model (FG), and Completely Randomized Design (CRD), alongside modified versions of ONA. These modified models, termed ONA.c and ONA.d, incorporated genetic relationships, specifically coancestry and genetic distance, instead of relying solely on clonal matching among adjacent trees. Subsequent cross-evaluation utilized four key indices - σ^2_{min} , d_{min} , FG index, and F_{min} - as suggested by ONA, minimum inbreeding, FG, and improved adaptive parallel genetic algorithm, respectively. The integrated process was applied through a case study involving the design of a *Larix kaempferi* seed orchard. Comparative analysis of indices revealed that ONA.c exhibited lower FG index and coancestry-related indicators, constituting 98.92% and 99.35% of the CRD and ONA indices, respectively. ONA.d demonstrated reduced F_{min} , a genetic similarity-related indicator, ranging from 85.66% to 86.52% compared to the other models. The modified ONA models show promise in mitigating inbreeding, particularly in advanced generations characterized by closer genetic relationships within a constrained gene pool. This integrated approach to seed orchard arrangement and evaluation holds potential for enhancing seed orchard establishment and management practices.

Keywords: seed orchards, inbreeding, genetic diversity, CRD, ONA

SEED ORCHARDS ARE KEY TO INCREASING RESOURCES FOR THE FUTURE

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Forests are fundamental for good livelihoods. They contribute to resilience of communities by regulating hydrology such as water flows, flooding and droughts. They stabilize soils and reduce erosion. They reduce impact of weather variations. They generate raw materials and income. They absorb carbon from the atmosphere and replace fossil carbon. A forest has some phenotypic plasticity, resilience and genetic diversity and can adapt by management. Planting or seeding can improve the forest for desired properties. Seed orchard seeds will add to those advantages. Seed orchard seeds (compared to stand seeds) are more predictable and reproducible. They constitute the most environmentally friendly way to increase forest production. Seed orchards contribute to an economically stronger forestry. Usually, they are physiologically better and cheaper to collect. Orchard trees typically origin from a wide range of stands and objects tested on a range of sites. This gives seeds suitable for variable conditions. The choice of orchard parents results in optimized genetic diversity and flexibility combined with other desired characters. Seed orchards create more renewable resources, which replaces fossil. Faster sequestering of carbon dioxide is a likely effect. Existing seed orchards can be modified to environments by rouging, selective harvest and changed target area. Human has an accelerating impact on environments, seed sources should adapt to that! Under climate change, the adaptability of local populations is replaced with assisted migration, where seed orchards are efficient. Clonal Seed Orchards with grafts of selected trees should be favored when: the situation is stable, long continuity in funding and seed demand, continuous scientific support and technical development, long-time experience with the species, good interaction of seed production, plant production and forestry, existence and access of suitable land. Seedling seed orchards are easier and cheaper to establish and manage. The risk of pathogens introduced will be reduced when using material from far away. Progeny tests can sometimes be converted to seedling seed orchards. For some situations, conversion of stands to seed sources may be the most realistic alternative, but if stands were not established with seed collection in mind, I do not call them seed orchards.

Keywords: seed orchards, resilience, genetic diversity, progeny tests

THE FIRST GRAYISH OAK SEED ORCHARD ESTABLISHED IN ROMANIA

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The marginal tree populations from the forest steppe are considered in danger due to the actual changes in the environmental conditions. One of the most valuable tree species with good potential for vegetation in the hard site conditions from the forest steppe is the grayish oak (*Quercus pedunculiflora atrichoclados* variety). The aim of this study was to establish the first grayish oak seed orchard in Romania, using vegetative propagations to ensure the production of adapted forest reproductive materials for the Dobrogea region. To obtain viable grafts, it was necessary to stimulate the annual branch development by carrying on high-intensity cuttings in the crowns of the selected graft donor trees/ramets, a year before. Collection of the grafts, grafting, monitoring seedlings, choosing the most suitable land and groundwork of the field and soil, setting up the seed orchard layout, and finally, establishing the seed orchard, were the main activities performed in this research project. Vegetative multiplication of grayish oak in a controlled environment, using the improved copulation, was the most efficient grafting method. Considering the total number of grafted seedlings and the clones' possible distribution on an area of 4.28 ha, a layout with 18 seedlings per row and 28 rows was used. In total, 504 grafted seedlings from 36 clones were planted in an incompletely balanced 6 × 6 grid design with 8 x 10 m spacing. Phenological observations on the graft's donor trees/ramets were made to compactly distribute the clones that disseminate the pollen in the same period. The Niculiţel grayish oak seed orchard was established in November 2017. In the autumn of 2023, at the age of 6 years, the seed orchard presented a good survey (>90%), high viability of ramets, and the first fruiting was recorded.

Keywords: breeding strategy, clone seed orchard, forest steppe, *Quercus pedunculiflora*, seed sources

AN OVERVIEW OF SEED ORCHARDS IN IRELAND

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The Forest Strategy for Ireland 2023-2030 places an emphasis on increasing the diversity of tree species established, including a significant increase in the use of broadleaf and native tree species. Under this strategy, The Irish Forestry Programme aims to increase the current land area of Ireland under forest cover (11% vs. EU average of 38%) whilst ensuring these climate resilient and healthy forests are managed according to the principles of sustainable forest management. The annual target for afforestation is 8000 ha. This presents challenges for FRM producers due to lead-in time to produce planting material (2-4 years depending on species), good seed years vary greatly by species and location, and storage of seed: some large-seeded broadleaves cannot be stored for any great length of time. To address these challenges, the Irish State is supporting (amongst other measures) the development of a seed orchard resource to increase the quantity and quality of forest tree seed. This poster gives an overview of seed orchards established in Ireland.

Keywords: Ireland's Forest Strategy, orchards, planting programme, afforestation, seed demand

GENETIC BACKGROUND OF EARLY FLOWERING IN BETULA PENDULA FOR THE SUCCESSFUL DEVELOPMENT OF EFFICIENT AND RAPID BREEDING

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Information on flowering genetics and flowering variability is important for the development of breeding programs. The importance of these will increase when breeding by means of genomic selection method will be implemented into operational breeding.

The project aimed to quantify the genetic parameters of early flowering in two genetic progeny trials located in southern Sweden. Both trials were planted in 2014 and consisted of offsprings from 19 full-sib families. The parents for the families came from the elite silver birch population for southern Sweden. There were around 20 - 70 plants per family in both trials. During three observation seasons, the occurrence of male and female flowers was registered. The number of flowers were quantified subjectively using a simple three stage scale from 0 (no flowers) to 3 (many flowers). Height, diameter, and quality traits were also collected.

Nine years after planting, there were around 20% of trees having male flowers. In one family, male flowering frequency exceeded 50%. The female flowering was much less frequent with an average less than 10%. Out of the 19 families, only 10 families had an individual with female flower. Flowering frequency was slightly higher at northernmost experiment and there was a positive correlation of transfer distance (distance between experiment location and parental origin) and male flowers production.

The male flowers started to be produced earlier than females' flowers. There was also a great genetic variation in ability to produce male and female flowers in early years after planting. Nine-years result also indicate a positive effect of northward transfer on frequency of trees producing male flowers. There is no such indication for female flower production. These results indicate that flowering ability must be considered in selection to facilitate performance of crosses in new generations and for a deployment of the results through seed-orchards. The indication of positive effect of transfer might be considered when new seed-orchards are planned for establishment.

Keywords: reproductive phenology, seed production

ASSEMBLY AND ANALYSIS OF CHLOROPLAST GENOME FOR DNA MARKER DEVELOPMENT IN QUERCUS ACUTISSIMA ‘GUMSURA 1HO’

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Quercus acutissima ‘Gumsura 1ho’, which have large acorn size and high yield, was developed from candidate *Q. acutissima* clones collected from nine locations in Republic of Korea. To develop discriminable DNA markers of ‘Gumsura 1ho’ among four *Quercus* (*Q. acutissima*, *Q. serrata*, *Q. mongolica*, and *Q. variabilis*), the chloroplast (cp) genome of ‘Gumsura 1ho’ was assembled. The cp genome size exhibited 161,151bp and revealed 134 genes including 86 protein-coding genes, 40 tRNA genes, and eight rRNA genes. We also found the insertion/deletions (indels) and single nucleotide polymorphism (SNP) between ‘Gumsura 1ho’ and reference cp genome (NC_039429.1) in *Q. acutissima*. When three indels and four cleaved amplified polymorphic sequences (CAPS) markers were developed to confirm sequence variants of ‘Gumsura 1ho’ cp genome using individual collected from four *Quercus* in seed orchard, PCR product size of ‘Gumsura 1ho’ was differed with individual of four *Quercus*. Subsequently, Taqman-assays were designed for high-throughput selection of ‘Gumsura 1ho’ in the confirmed two SNP regions, and then ‘Gumsura 1ho’ was discriminated among four *Quercus*. Therefore, our results suggested that developed DNA markers could be used for discrimination of ‘Gumsura 1ho’ clone among four *Quercus*.

Keywords: chloroplast genome', 'DNA marker', 'genome assembly', 'quercus acutissima'.

THE NEWEST SEED ORCHARD OF *PICEA ABIES PENDULA* FORM ESTABLISHED IN ROMANIA

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The main goal of promoting the narrow-crowned Norway spruce is related to its superiority compared to the normal crown form (pyramidal), expressed by larger dimensions, superior wood quality, and better resistance to natural disturbances. The aim of the research project was the ex-situ conservation simultaneously with the production of forest reproductive materials by cloning the last individuals of the narrow-crowned Norway spruce (*Picea abies f. pendula*). 29 pendula spruce clones were selected from three natural populations (Stâna de Vale and Iz buc, from the Apuseni mountains, and Predeal, from the Curvature Carpathians) and one seed orchard (Soveja) to harvest the necessary grafts. One year before grafting, spruce rootstock seedlings of 2-3 years old, with a minimum thickness of 7 mm, straight stems, without defects, and a distance of a minimum of 5 cm among whorls, were planted in polyethylene bags. The grafting method used was side-veneering and involved the following activities: preparation of the grafts and compatible rootstock seedlings, tight bonding of the two symbionts, covering the grafting area with wax, labeling, and separate storage of clones. The grafted seedlings were monitored for one growing season in an air-conditioned solarium, and later in the nursery (performing all activities for optimal development). Vegetative multiplication by grafting had a success rate of 37%, resulting in 661 grafted seedlings related to 25 clones. The Săcele seed orchard of Norway spruce *pendula* form was established in the spring of 2022 (May), with 506 grafted seedlings reported to 25 clones in a 5 x 5 grid design and at a 4 x 4 m distance among seedlings. Monitoring of the seed orchard will require a whole series of actions, staggered over several years, respectively: additions, irrigation, phytosanitary treatments, fertilizers, clearing, and maintenance of the fence.

Keywords: breeding strategy, forest reproductive materials, ex-situ conservation, ideotype trees, Norway spruce

CHALLENGES FOR IMPLEMENTING ASSISTED MIGRATION INTO A SEED ORCHARD PROGRAMME IN SAXONY, GERMANY

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The Public Enterprise Sachsenforst (SBS) is implementing a 5-year seed orchard (s.o.) programme that includes the maintenance of existing, but also the establishment of new s.o. The focus for new s.o. is laid on indigenous broadleaf species that are expected to improve the species diversity and the climate (mainly drought) resilience of the future Saxon forests. However, a recent expert opinion that relied on extrapolation models predicting the regional change of climatic site conditions in Saxony has revealed that, already today, new target forest types, which should guide the operational decisions, have to be determined for silvicultural practice in Saxon forests. The predicted target forest types comprise species that are new to Saxony and native to southern Europe, such as *Castanea sataiva*. However, also, the species that are native to Saxony today should be assisted in their migration by introducing provenances from Southeast Central and South Europe to Saxony. The presentation will explain the theoretical questions and practical challenges for present-day seed orchard establishment in Saxony. For instance, uncertainty exists with the need for climate change adaptation of the population genetic structure of main indigenous species, such as the white oaks *Quercus petraea* or *Q. robur*. There is a lack of knowledge regarding the distance of sourcing zones from Saxony for assisted migration along the post-Pleistocene re-invasion pathways. In addition, the extent of assisted upward elevation shifts of lowland provenances into the low mountain range of Saxony has to be considered under the increasing risk for late-frost incidents while the vegetation period has become longer. For the common secondary species, such as hornbeam or Norway maple, still today, population genetic structures are often poorly understood because spatial DNA markers, which allow drawing the bio-geographic boundary zones, cannot be gathered. The population density of these species is too weak in Saxony and potentially also in the neighbouring countries.

Keywords: climate resilience; species composition; Hercynian mixed mountain forest; broadleaf; heterozygosity; plus-tree origin

STUDY OF THE DECLINE OF CONIFEROUS FRUITING IN THE LANDES MASSIF

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The importance of the seeds and seedlings sector for maritime pine (*Pinus pinaster*) but also the strong dynamic of varietal creation make it necessary to master the management of seed orchards in order to improve the availability of seeds of high genetic value. Since 2009, massive dieback of maritime pine conelets in the first year fruiting and a low yield of seed cones are noted on maritime pine orchards for several years. We also observed many seeds which are not viable. Different causes could be at the origin of this damage: bio-aggressors such as bugs *Leptoglossus occidentalis*, climate change influencing phenology flowering or the incidence of late frosts, low site fertility establishment of orchards. *Leptoglossus occidentalis*, known in the United-State and officially observed in France in 2007 (Dusoulier et al. 2007), seems to bite cones. For these reasons, we installed some traps and try lures to know what happened by who and when on three different resinous trees (*Pinus pinaster*, *Pinus taeda*, Douglas fir). Following the questioning of the different treatments tested previously, it is necessary to review the current health protection plan to find ways of improvement (choice of product, dose, period of application) and look for new control strategies. The results of the direct microinjection trial in the trunk are encouraging and we continue to work on it to find a solution of this loss.

Keywords: seeds production, *Pinus*, traps, *Leptoglossus occidentalis*, fructification

FLOWERING AND SEED PRODUCTION ON 20-YEAR-OLD SILVER BIRCH CLONE SEED ORCHARD IN CENTRAL POLAND

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Silver birch (*Betula pendula* Roth) is a commonly occurred deciduous forest-forming species in Poland, covering approximately 7% of the forest area. In Poland, for silver birch 10 seed regions of origin have been established, including 2 of distinguishing phenotypical quality. The production of genetically improved seeds of birch takes place on first-generation seed plantations: 12 Seed Orchards (SO) and 2 Seedling Seed Orchards (SSO), with a total area of 65 ha, and 14 ha, respectively. The production capacity of seeds depends on many factors, and it varies (from 3 to 120 kg), with an average amount of 50 kg of infructescences per 1 ha.

The aim of study was determination of the variability of flowering and seed productivity, on the SO in central Poland (51.5700, 20.2681), on an area of 9 ha with 1506 grafts from 32 clones, and 6 x 6 m spacing of planting density. The assessment of the development of male flowers, and the intensity of yielding, and the stage of maturity of infructescences were performed once using a scale from 0 (none) to 5 (abundant). Then, one of the trees from harvesting classes 3, 4, and 5 were cut down, and all seeds were collected to assess, their field volume, weight, and moisture.

Three clones (7827, 7838, 7843) stand out with the best flowering - 5-8% of the grafts showed abundant rate; the worst were 4 of them (7824, 7830, 7832, 7841), where more than 8-10% of the grafts did not produce anthers at all. The best-yielding clones, whose percentage share in the production of seeds of the entire seed plantation is 5.1-6.0%, include 7838, 7825, 7827, and 7819. On average, each grafts of these clones produced 2.5-2.9 kg of seeds. The lowest-yielding clones 7834, 7841, 7842, 7832, and 7830 ensure seed production at only 0.7-1.5% of the entire seed plantation, while the average production of seeds is estimated at 0.3-0.7 kg. The calculated total yield of infructescences, based on the yielding classes of all grafts per 1 ha of seed plantation is 191 kg.

Keywords: seed orchards, silver birch, flowering, seed productivity, yielding