



NIBIO
NORWEGIAN INSTITUTE OF
BIOECONOMY RESEARCH

NIBIO BOOK | VOL. 1 NR 01 2015

The 12th International

Christmas Tree Research and Extension Conference

Honne, Norway

6th – 11th September 2015



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V. Talgø & I. S. Fløistad, eds.





The publisher of NIBIO BOOK is:
NIBIO, P.o. box 115, N-1431 Ås, Norway
post@nibio.no

Editor in charge: Director of Research Nils Vagstad
Current issue: NIBIO Plant Health and Biotechnology Division
Local editor: Dr. Agric Dan Aamlid
Scientific editors: Venche Talgø & Inger Sundheim Fløistad
Design: Miksmaster Creative

NIBIO BOOK
Vol 1 nr 1 2015
ISBN: 978-82-17-01453-9
ISSN ISSN 2464-1189

Photo: V. Talgø: p 8-9, 14-15, 51, 52-53.
I. S. Fløistad: Cover, p 4.

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Dear Christmas tree friends

It is our pleasure to welcome you to Norway and the 12th International Christmas Tree Research and Extension Conference. In total, there are about 40 participants from Canada, USA, Australia, Austria, Iceland, Greece, Hungary, UK, Denmark, France and of course Norway.

Our meeting will be opened by the director of NIBIO Plant Health Division, Dr. Arne Hermansen. We are very grateful for the support he has given us in organizing the conference, and for taking the time to travel to Honne to welcome you and give an introduction to the new institution NIBIO, Norwegian Institute of Economy Research, which was established 1 July 2015 as a merger between the Norwegian Institute for Agricultural and Environmental Research (Bioforsk), Norwegian Forest and Landscape Institute and the Norwegian Agricultural Economics Research Institute.

We are also very grateful to the hosts of the fieldtrips. Dr. Knut Huse will guide us through the local arboretum at Honne on Monday. On Tuesday, we are welcome to visit the forest nursery at Biri, Skogplanter Østnorge AS. Thursday is entirely dedicated to visits to Christmas tree growers, a subalpine fir provenance trial and a seed orchard. We are most grateful to John-Anders Strande, general manager of The Norwegian Christmas tree grower association, who has facilitated the excursion on Thursday. Finally, on Friday, we are welcome for a guided tour through the production at the Norwegian Forest Seed Center.

Furthermore, we want to thank for the support given by our Director of research Nils Vagstad, who introduced the idea of publishing presentations in Scandinavian Journal of Forest Research after the conference. All technical assistance from Kari Munthe and Erling Fløistad is also highly appreciated.

During the conference, the following topics will be covered:

- Breeding & genetics
- Insects
- Tree health
- Physiology
- Growth conditions & integrated pest management
- Postharvest, market & economy

Abstracts of oral presentations are printed in the order they appear in the program. Poster abstracts follow after the oral abstracts, and are not organized in topics.

We hope this will be an interesting conference and a good opportunity for the participants from different countries to exchange results and discuss challenges regarding Christmas tree production.

**Venche Talgø and Inger Sundheim Fløistad,
Organizing committee**

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PROGRAM



The 12th International

Christmas Tree Research and Extension Conference

Honne, Norway

6th – 11th September 2015

Sunday 6th September

- 16.00 Registration and poster setup
19.00 Welcome reception/dinner

Monday 7th September

09.30 OPENING SESSION

Opening of the Conference
Arne Hermansen, Director of NIBIO Plant
health division

Producing Christmas tree in
“the land of the midnight sun”
John Anders Strande

- 10.05 BREAK

SESSION 1: BREEDING AND GENETIC

- 10.30 **Environmental control of cone production in Fraser fir (*Abies fraseri*) Christmas trees**
Brent Crain, Pascal Nzokou, Jill O’Donnell,
Beth Bishop & Bert Cregg
- 10.50 **Comparing noble fir progeny from collection regions in the Pacific Northwest and Denmark**
Chal Landgren, Ulrik Braüner Nielsen & Gary Chastagner
- 11.10 **The use of plant growth regulators for coning and height control in *Abies* and *Picea***
Brent Crain & Bert Cregg
- 11.30 LUNSJ
- 12.30 **Field trial with *Abies lasiocarpa* progenies for Christmas tree production in Norway**
Inger Sundheim Fløistad, Hans Nyeggen &
Jan-Ole Skage
- 12.50 **Genomic technologies and applied tree breeding**
Ross Whetten
- 13.10 **Guided walk in the arboretum at The Forestry Extension Institute at Honne**
Knut Huse

SESSION 2: INSECTS

- 14.50 **Risk of *Adelges (Dreyfusia) nordmanniana* surviving on cut Nordmann fir Christmas trees and boughs**
Gary Chastagner, Kathy Riley & Andy McReynolds
- 15.10 **Effect of Bornyl Acetate on Reproduction of the Green Peach Aphid and Balsam Woolly Adelgid**
Ethan Bucholz, David Tilotta, Robert Jetton,
Lucian Lucia & John Frampton
- 15.30 **Evaluating Beneficial Insects for Aphid Control in Christmas Trees**
Chal Landgren, Jana Lee, Judi Kowalski &
Ryan Hill
- 15.50 BREAK
- 16.20 **Review of new insecticides examined in Denmark for control of silver fir woolly aphid (*Dreyfusia nordmanniana*) in Christmas trees**
Paul Christensen
- 16.40 **Insects causing plant protection problems on Christmas tree plantations in Hungary**
Katalin Tuba & Géza Kelemen
- 19.00 DINNER

Tuesday 8th September

SESSION 3: TREE HEALTH

- 09.00** **Increased number of *Phytophthora* species found in Fraser fir Christmas Tree Plantations in the Southern Appalachians**
Jan Martin Pettersson, John Frampton, Jonas Rönnberg, David Shew, D. Michael Benson & Marc A. Cubeta
- 9.20** **Response of Turkish and Trojan Fir Seedlings to *Phytophthora cinnamomi* and *P. cryptogea***
Will Kohlway, Ross Whetten, D. Michael Benson, Anne Margaret Braham & John Frampton
- 9.40** **Mapping *Phytophthora* Root Rot Resistance in Fir Species through Genotyping by Sequencing**
Will Kohlway, John Frampton & Ross Whetten
- 10.00 BREAK
- 10.30** **Screening *Abies* for Resistance to *Phytophthora* Root Rot**
Kathleen McKeever & Gary Chastagner
- 10.50** **Fungicide resistant *Botrytis* strains are present in forest nurseries in Norway**
Gunn Mari Strømeng, Venche Talgø & Inger Sundheim Fløistad
- 11.10** **Identification and pathogenicity of *Phomopsis* isolates associated with spruce decline in Christmas and landscape tree settings in Michigan, USA**
Christine K. McTavish, Mursel Catal, Jill O'Donnell, Dennis W. Fulbright & Andrew M. Jarosz
- 11.30 LUNCH
- 12.30** **Fieldtrip to a forest nursery Skogplanter Østnorge AS, Biri**

SESSION 3 TREE HEALTH, CONTINUED

- 15.00** ***Neonectria neomacrospora* has caused severe damage on true fir (*Abies* spp.) in Denmark**
Iben Margrete Thomsen & Venche Talgø
- 15.20** **Development and application of a PCR-based test for the identification of *Neonectria neomacrospora* damaging *Abies* species**
May Bente Brurberg, Arne Stensvand & Venche Talgø
- 15.40** **Inoculation experiments with *Neonectria neomacrospora* on *Abies nordmanniana***
Iben Margrete Thomsen, Jing Xu & Venche Talgø
- 16.00** ***Neonectria* – an update on genetic variation in tree susceptibility based on ocular field evaluations**
Ulrik Brauner Nielsen, Jing Xu, Venche Talgø & Iben M. Thomsen
- 16.20 BREAK
- 16.50** **Seed-borne fungi on Christmas trees**
Guro Brodal, Heidi Røsok Bye, Eleonora Høst & Venche Talgø
- 17.10** **Does the severity of current season needle necrosis decrease on older stands of noble fir?**
Gary Chastagner, Chal Landgren & Ulrik Brauner Nielsen
- 19.00 DINNER

Wednesday 9th September

POSTER SESSION

Short presentation and questions at the display site
(approximately 5 min. per poster)

10.00 BREAK

SESSION 4: PHYSIOLOGY

10.30 Does temperature at the developing bud regulate cone production in *Abies fraseri*?
Brent Crain & Bert Cregg

10.50 Variation in Survival and Bud Break of Turkish and Trojan fir in the United States
Yusuf Kurt, Chal Landgren, Gary Chastagner, Bert Cregg, Pascal Nzokou, Jill O'Donnell, Rick Bates, Ulrik Bräuner Nielsen, Richard Cowles, Fikret Isik & John Frampton

11.10 Using Herbicides to Interrupt Cone Development on Fraser Fir
Jeffrey Owen

11.30 LUNCH

SESSION 5: GROWTH CONDITIONS AND IPM

12.30 Evaluating nitrogen source and application timing for optimal nitrogen uptake
Jill O'Donnell, Bert Cregg, & Erin Lizotte

12.50 Integrated Pest Management Education for the Christmas Tree Industry in Oregon, USA
Luisa Santamaria & Chal Landgren

13.10 Management of diseases in Norwegian Christmas tree fields
Venche Talgø & Inger Sundheim Fløistad

13.30 BREAK

SESSION 6: POSTHARVEST, MARKET AND ECONOMY

14.00 Postharvest moisture status and quality of trees displayed in tenon-type Christmas tree stands
Gary Chastagner & Andy McReynolds

14.20 Inferring gene networks regulating needle abscission in Fraser fir through RNA-seq data
Lilian Matallana, Kathryn Coats, Gary Chastagner, John Frampton & Ross Whetten,

14.40 Seasonal changes in balsam fir needle abscission patterns and links to environmental factors
Mason T. MacDonald & Rajasekaran R. Lada

15.00 BREAK

15.30 The European Christmas Tree industry – aspects of markets and production
Claus Jerram Christensen

15.50 The challenges of establishing a niche market for CHO.C.O. (CHOose, Cut, Offset) Christmas Trees farms in Greece
Konstantinos G. Paspapryopoulos & Nikolaos Grigoriadis

16.10 FjordTree
John-Anders Strande

19.00 DINNER

Thursday 10th September

08.00 Fieldtrip, including dinner

Friday 11th September

08.00 Business meeting
09.30 Bus leaving for fieldtrip
12.00 Bus leaving The Norwegian Forest Seed Center
13.15 Arrival at Gardermoen airport





**ABSTRACTS
ORAL PRESENTATIONS**

OPENING SESSION

Producing Christmas trees in “the land of the midnight sun”

J.-A. Strande

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“Norsk Juletre” is the Norwegian Christmas tree grower association with 470 members and 7 local associations. The association is a member of CTGCE (Christmas Tree Grower Council Europe). Christmas tree production is a relatively new agricultural concept in Norway. From the tradition of keeping a tree for Christmas came to Norway around 1825 and until the nineteen eighties, trees have traditionally been taken from the forests as a thinning, early in the rotation. Although this still goes on today, fewer seem to find the time for such activities.

Christmas tree production became popular in the last part of the nineteen nineties, and a large amount of non-rational agricultural land was planted. Many of those who planted in this period were under the impression that the job was done when the plant was in the ground. Many of these plantations, which lacked the demanded quality, were sold off on export after the millennium and reclaimed for pastures and so on. Some of them are today dense forests. This again have resulted in a scepticism from agricultural authorities towards accepting planting on agricultural land, and scepticism from landowners towards committing to produce Christmas trees.

The early professional producers orientated themselves towards Denmark to gain knowledge. Therefore Nordmann fir (*Abies nordmanniana*) became a popular tree, especially in the south-west where the climate allowed this production. Subalpine fir (*A. lasiocarpa*) then became demanded on the market and now constitutes about 50% of the fir market. Although the south-west still is an important area for Norwegian Christmas tree production, subalpine fir allows production of fir over most of the southern half of Norway.

Annual consumption of natural trees in Norway is 1.900.000 trees. Annual Import is 325.000 trees (+unregistered import). Trees annually taken out of conventional forestry are 400.000 trees and annual professional produced trees in Norway is 1.100.000 trees. The annual national market of natural trees is 60% fir (50% Nordmann fir and 50% subalpine fir) and 40% spruce, mainly Norway spruce (*Picea abies*). Totally, 20% of consumer say they use an artificial Christmas tree (numbers from 2012 and 2013). Prices from 2014 were NOK 450 (EUR 54, USD 60) for spruce and NOK 600 (EUR 72, USD 80) for fir.

SESSION 1: BREEDING AND GENETIC

Environmental control of cone production in Fraser fir (*Abies fraseri*) Christmas trees

B. Crain¹, P. Nzokou², J. O'Donnell³, B. Bishop⁴ & B. Cregg^{1,2}

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Due to its beautiful form, strong branches, and exceptional needle-retention, Fraser fir is rapidly gaining in popularity as a Christmas tree in Michigan and elsewhere in the United States. Michigan plantation-grown Fraser fir trees often produce heavy cone crops at a much younger age than those in natural stands. Cones must be removed by hand at considerable expense, and cone production alters the tree structure, decreasing the value as a Christmas tree.

In true firs, cone buds differentiate during the summer, but do not open until the following spring. Thus environmental conditions during summer bud differentiation may regulate cone production for the following year. Fraser fir is endemic to a small region of cool temperate rain forest in the southeastern Appalachians. Annual precipitation is high—twice that of central Michigan—and mean summer temperatures are below 16°C, which is much cooler than Michigan. It seems likely that these climatic differences play a significant role in heavy coning in Michigan. This is consistent with what is known about conifer reproduction in general, much of which comes from research to promote cone production in seed orchards: Tree age, size, hormonal interactions, water and nutrient availability, and temperature are key drivers of cone development (Owens and Blake 1985; Owens 1995). In addition, we have observed dramatic variation in coning within individual fields of singular seed source and planting date (Crain *et al.* 2012). This suggests that highly localized environmental signaling regulates cone development.

In 2011, we established multiyear observational studies to understand the environmental factors regulating early cone development. Using multiple regression on data collected from 10 farms over 4 years, we are developing models to predict cone crop yield based on weather conditions during the preceding summer. This may provide advance warning to growers, giving them time to procure extra workers to remove cones in particularly heavy coning years.

In 2013, we established designed experiments at multiple locations to examine the effects of environmental variables (water, temperature, nutrition) on cone production. In our mulching and irrigation studies, results suggest that drought and heat stress increase coning, but that neither irrigation nor mulching sufficiently reduce stress to control coning.

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- Crain, B., Cregg, B., Nzokou, P., O'Donnell, J. & Bishop, B. 2012. Factors affecting early cone development in Fraser fir. *HortScience*. 47:234.
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Comparing noble fir progeny from collection regions in the Pacific Northwest and Denmark

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Noble fir (*Abies procera* Rehd.) is the leading Christmas tree produced in the states of Oregon and Washington (USA) with yearly plantings of over 4 million trees. In Denmark, noble fir is primarily utilized for greenery, though Christmas tree production is expanding. Progeny and provenance testing utilizing commercial plantations has been ongoing since mid-1970's. Testing since 1996 has included 215 families (primarily 1/2 sibling families) from across the natural range of noble fir and includes families from Danish imported seed. Traits investigated for Christmas trees include height, grade, color and incidence of Current Season Needle Necrosis (CSNN). The 6 regions that are compared and the number of observed progeny are:

- Oregon Cascades (936 observed trees) - From Mt. Hood in the north to the McKenzie River in the south, noble is fairly continuous in distribution beginning at elevations above 3,500 feet.
- Oregon Coast (13,178 observed trees) - The distribution is scattered on isolated peaks above 2000 ft.
- S. Or Cascades (1,412 observed trees) - This is where noble mixes with Shasta fir in the area of the McKenzie River at elevations above 3000 feet.
- Washington Cascades (1695 observed trees) - The distribution begins at Larch Mountain in the south and extends to Stevens Pass where the natural distribution ends.

- Willapa Hills, Washington (564 observed trees) - Many suggest this area is an extension of the noble fir distribution of the Oregon Coast. Noble fir is found only on the upper elevations of a few mountain peaks, notably BawFaw/ Boistfort.
- Danish Collections (1199 observed trees) - All of the Danish collections originated from the PNW. Likely, collection sites are in on Mt. Hood and perhaps in the Washington Cascades out of Fort Vancouver. Selection and breeding have focused on traits for greenery over many years.

Evaluations suggest the 1/2 sibling sources from the coastal mountains in Oregon, consistently are among the top for Oregon and Washington producers based on tree value. Selections from the Oregon and Washington Cascade mountains consistently rank lower for value. Sources from the southern limit of noble fir in the Cascades are consistently slower growing with an open growth habit and share traits with Shasta fir (*A. magnifica* var. *shastensis*). The tested Danish sources share many traits with the Cascade mountain sources with consistently high evaluations for superior color and low CSNN incidence.

The use of plant growth regulators for coning and height control in *Abies* and *Picea*

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Early efforts to increase cone production in seed orchards relied on cultural treatments, such as girdling, root pruning, fertilization, and induced drought (Puritch 1972). Results were highly variable, and occasionally detrimental. Subsequent work with plant growth regulators (PGRs)—particularly exogenous application of gibberellins (GAs)—resulted in greatly enhanced cone production, especially when combined with cultural treatments (Puritch 1979). Little research has been directed toward the use of PGRs to regulate cone production in *Abies*, but trunk-injection of GA_{4/7} combined with girdling and tenting does increase cone production in *Abies amabilis* (Pacific silver fir; Owens *et al.* 2001).

In Michigan plantations, Fraser fir (*A. fraseri*) Christmas trees frequently produce heavy cone crops, which are expensive to remove and may reduce the value of the tree. Since GAs are often used to enhance coning in conifer seed orchards, it seems reasonable that GA-inhibitors may reduce cone production. GAs are also involved in stem elongation, so GA-inhibitors should reduce vegetative growth and may decrease the need for shearing. In 2013, we established four-year studies at four locations to evaluate the use of PGR treatments for coning and height control in Fraser fir. Twenty trees were randomly assigned to one of five treatments: 1) water control; 2) Provide (GA_{4/7}), positive control; 3) Cycocel (chlormequat); 4) Trimtect (paclobutrazol); 5) Cambistat (paclobutra-

zol). Treatments 3–5 are GA-biosynthesis inhibitors. Treatment 5 was applied one time in early spring by soil injection. All other treatments were applied 3 times at 10 day intervals by foliar drench, during the period of cone bud initiation and differentiation. In the first year, GA inhibitors reduced cone production by 15–70%, and GA doubled cone production. However, results were not consistent across sites.

We also established studies at multiple locations to evaluate the use of PGRs in height control in blue spruce (*Picea pungens*) and Norway spruce (*P. abies*). In spring 2013, 20 randomly selected trees were treated once with Cambistat by soil injection, and 20 trees were selected as untreated controls. Trees were scored each fall for bud density and height and lateral growth. Height control was highly significant in 2013 and 2014. 2014 results from one site in central Michigan were typical, with average leader growth of 24.3 cm for treated small blue spruce, compared with 36.7 cm for the control—a difference of 40.5%.

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Field trial with *Abies lasiocarpa* progenies for Christmas tree production in Norway

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Abies lasiocarpa (Hook.) Nutt is together with *A. nordmanniana* (Steven) Spach the most common exotic species for Christmas tree production in Norway. While *A. nordmanniana* is used mostly in coastal areas, *A. lasiocarpa* can also be grown in the interior part of the country where the climate generally is too harsh for growing *A. nordmanniana*.

The wide natural range of *A. lasiocarpa* in western North America, from Alaska and Yukon in the north to Arizona and New Mexico in the south, represent great differences among populations in growth, morphology and phenology. Provenance field trials in Norway for Christmas tree production have previously been performed by Hansen *et al.* (2004) and Skage *et al.* (2012). However, progenies of *A. lasiocarpa* from seed orchards and from seed collected from plus trees have never been compared in trials in Norway. Two experimental plots were established in the southern part of Norway for testing progenies from selected plus trees and from seed orchard clones.

The experiment included progenies from 17 selected plus trees with superior morphological traits from 11 provenances in US and Canada (Stavrum & Johnskås 1995, Stavrum & Gislerud 1996), and progenies from 6 seed orchard clones in Kaupanger in the western part of Norway. One experimental site was established in the spring of 2000, in Luster municipality, Sogn og Fjordane County in western Norway. The other site was established in the spring of 2001 in Stange municipality, Hedmark County in eastern Norway.

The average survival rate was 71% in Luster and 74% in Stange, with the highest mortality during the first three years. Provenances from Wyoming appeared with low survival and Christmas tree yield in Luster. In Stange, a high number of trees were damaged by frost in late spring 2008, however with less frost damages in provenances from Wyoming and Alberta.

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Genomic technologies and applied tree breeding

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The past ten years have seen phenomenal progress in the development of tools for detecting and analyzing genetic and biochemical variation. These tools have generally been developed and first applied in biomedical research, and have then spread to other fields as diverse as agriculture, ecology, population biology, and forestry. These tools are sometimes referred to as genomic technologies, because they allow analysis of many, if not all, genes or gene products in an organism in parallel. Such methods have been applied in research projects on forest trees over the past decade, and the question naturally arises of when they might find application in practical breeding programs working with Christmas tree species. This presentation will provide an overview of genomic technologies, including high-throughput methods for discovery and analysis of genetic variation as well as methods for detection of regulatory interactions among genes or between genes and environmental signals. Some examples of research projects un-

derway using these methods will be described, and future prospects for integration of these methods into applied breeding programs will be discussed. A key question is how to decide when these technologies are ready to move into application in practical breeding programs, and a reasonable approach is to prioritize the opportunities based on the probability of return on investment and the opportunity cost of failing to apply tools as they become available.

SESSION 2: INSECTS

Risk of *Adelges (Dreyfusia) nordmanniana* surviving on cut Nordmann fir Christmas trees and boughs

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In Europe where Nordmann fir [*Abies nordmanniana* (Steven) Spach] is widely grown for Christmas trees and boughs, the silver fir woolly adelgid [*Adelges (Dreyfusia) nordmanniana*] is a serious pest on this host. Although not common, this pest has been observed on Nordmann fir trees at several locations in western Washington. During the past few years, data has been collected on its rate of spread and life cycle in plantings at Puyallup. Information about host susceptibility and the effectiveness of insecticide treatments in controlling this pest have also been collected.

In an effort to determine the risk that adelgids could be spread from one location to another via the movement of infested cut Christmas trees or boughs, experiments were done in 2013 and 2014 to examine the potential for adelgids to survive on harvested boughs. Branches from five heavily-infested Nordmann fir trees were utilized during this test. Three sets of branches, consisting of a single branch from each tree, were harvested in December/January. One set was stored in ventilated plastic crates outdoors. The remaining two sets were displayed indoors at 20°C for about 5 weeks. One set of the displayed branches was displayed with their bases in water and the other set was displayed dry. Following the indoor display period, both sets of the displayed branches were placed in ventilated plastic crates and stored outdoors with the other branches. Checks consisted of branches that were tagged, but not harvested from the tree. The effect of these different display and storage conditions on adelgid survival was determined by periodically examining the branches to determine the viability and life stages of the adelgids through early April.

There was no evidence of mortality of the overwintering adelgids on the unharvested branches on the trees. They started laying eggs in March and crawlers were evident by early April, which was about 3 weeks prior to bud break. In 2013, the adelgids on the harvested branches that were displayed indoors in water laid eggs which hatched, producing crawlers during the indoor display period. By the end of the display period, there was no evidence of live stem mother adelgids, eggs or crawlers on any of the branches that were displayed dry. No eggs were ever found on the branches that were originally cut and stored outdoors. By mid-March to early April, there were no surviving adelgids on any of the harvested branches, suggesting that there is virtually no risk of spreading the silver fir woolly adelgid from one area to another via cut trees or boughs.

Effect of Bornyl Acetate on Reproduction of the Green Peach Aphid and Balsam Woolly Adelgid

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Fraser fir (*Abies fraseri* [Pursch] Poir.) Christmas trees are an important crop in the Southern Appalachian region of the United States with an annual revenue exceeding \$US 100 million in North Carolina alone. Although most growers in the region utilize Integrated Pest Management (IPM) practices, the exotic balsam woolly adelgid (BWA, *Adelges piceae* Ratzeburg) forces growers to employ expensive insecticidal treatments at a cost of over \$US 1.5 million annually to maintain the marketability of their crop. An understanding of the chemical basis for BWA resistance is therefore essential to accelerate development and deployment of resistant planting stock and mitigate the impact of this destructive pest.

Although fir species are known to vary in their resistance to BWA, there is little evidence to suggest why some species such as Fraser fir are very susceptible, while others such as Veitch fir (*A. veitchii* Lindl.) from Japan are highly resistant. Our profile comparisons of extractives (acetone-soluble) from the stem of Veitch and Fraser fir tissue via gas chromatography coupled with mass spectroscopy (GC-MS) have consistently shown markedly higher amounts of bornyl acetate (BA) in Veitch fir. As a first step toward evaluating the effect, if any, of BA on the life cycle of BWA, the objective of this study is to determine the effect of various BA concentrations in the headspace above egg masses and adults on egg eclosion in BWA and vivipary (live birth) of a surrogate, the green peach aphid (*Myzus persicae* Sulzer).

We have developed a very simple protocol to vary the volatile BA concentration in the headspace of vessels that consists of diluting BA in silicone oil. Concentrations are measured by solid phase micro-extraction (SPME) fiber collection followed by GC-MS injection and analysis. In separate trials, adults of green peach aphid or eggs of BWA are placed into vessels with 5 different concentrations of BA (bracketing those measured for Veitch and Fraser fir) in addition to control vessels containing only silicone oil or water. After a week, samples are frozen and eggs and insects examined microscopically. The effect of BA on reproduction will be presented.

Evaluating Beneficial Insects for Aphid Control in Christmas Trees

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The past ten years have seen phenomenal progress in the development of tools for detecting and analyzing genetic and biochemical variation. These tools have generally been developed and first applied in biomedical research, and have then spread to other fields as diverse as agriculture, ecology, population biology, and forestry. These tools are sometimes referred to as genomic technologies, because they allow analysis of many, if not all, genes or gene products in an organism in parallel. Such methods have been applied in research projects on forest trees over the past decade, and the question naturally arises of when they might find application in practical breeding programs working with Christmas tree species. This presentation will provide an overview of genomic technologies, including high-throughput methods

for discovery and analysis of genetic variation as well as methods for detection of regulatory interactions among genes or between genes and environmental signals. Some examples of research projects underway using these methods will be described, and future prospects for integration of these methods into applied breeding programs will be discussed. A key question is how to decide when these technologies are ready to move into application in practical breeding programs, and a reasonable approach is to prioritize the opportunities based on the probability of return on investment and the opportunity cost of failing to apply tools as they become available.

Review of new insecticides examined in Denmark for control of silver fir woolly aphid (*Dreyfusia nordmanniana*) in Christmas trees

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In Denmark, as well as most other EU-countries (European Union), there is increased concern of the environmental effect caused by usage of pesticides. Especially insecticides that are very harmful for water living organisms make politicians and other people concerned. The traditionally used insecticides for Christmas trees, synthetic pyrethroids, were more or less banned in Denmark for usage in Christmas tree production from 2012. At present only a reduced dose of lambda-cyhalothrin (trade name Karate) is allowed in Christmas trees, but the dose is too low to give certainty for sufficient aphid control. Finally a dramatical increase in taxing was put on pyrethroids, so pine weevil control in forestry and other insect control in agriculture is now very costly.

During the last years a lot of new insecticides therefore have been tested in Denmark. One of them has now been registered via a minor use registration for use against various aphid species.

Some of the new insecticides have been tested with addition of additives, but this did not give better effect to the aphids, while it significant increased the risk of discoloration of the needles when applied after bud break.

Among the newly tested insecticides, until now only acetamiprid (trade name Mospilan) has been registered for use in Christmas trees in Denmark.

Acetamiprid (with trade name Mospilan) is one of several neo-nicotinoides which gives, as most other newly tested insecticides, a very good effect to aphids just like the traditionally used pyrethroids. Acetamiprid has been reported to be systemic in some agricultural crops, but this effect could not be observed in coniferous tree species.

Acetamiprid has not been a cause to discoloration of Nordmann fir (*Abies nordmanniana*) even if the buds had broken.

It seems to be a never ending work to test and get new insecticides registered, and to find other pesticides for replacement instead of pesticides banned by the authorities.

Insects causing plant protection problems on Christmas tree plantations in Hungary

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Eight to ten species of four coniferous tree genera (*Abies* spp., *Picea* spp., *Pinus* spp., *Pseudotsuga* spp.) are cultivated on the Hungarian Christmas tree plantations nowadays. Only a smaller part of our country is suitable to cultivate these species but in this area it provides a substantial source of income for rural dwellers.

Integrated plant protection plays a dominant role in the effective and economical Christmas tree cultivation. In this culture the aesthetic expectations are high but 'De gustibus non est disputandum' so plant protection tries to support these priorities, too. The susceptibility of cultivars, the presence of some invasive species, furthermore the spread and propagation of some species, due to the climate change cause serious problems on these plantations.

The timing of the chemical treatments is difficult. This culture can be characterised by a lack of lures and trapping methods. Due to these conditions it is typical that the farmers overuse the chemical plant protection methods.

The most harmful insects belong to the suborder Sternorrhyncha. In most cases firs are infested by aphid species such as *Dreyfusia* and *Cinara* species.

Sacchiphantes and *Adelges* gall maker aphids determine the growth of the Norway spruce in Hungary. *Physokermes* species also cause a serious problem, especially at the blue spruce. *Epinotia* species and *Cydia pactolana* may become dangerous on certain plantations. They can also support the secondary infection of the fungus.

Black pine is a unique Christmas tree species in Hungary. This species has a good resistance against insects. However, there are some years when sawfly species give a headache to plant protection professionals.

Douglas fir is endangered by a mealybug species (*Gillettella cooley*).

The protection against the above mentioned species is difficult due to their morphology, life-cycle and their shelters.

As a special case the invasive harlequin ladybird can be mentioned, which would like to overwinter in the Christmas trees. They are sprayed in autumn because they cause inconveniences to the customers of Christmas trees

SESSION 3: TREE HEALTH

Increased number of *Phytophthora* Species found in Fraser fir Christmas Tree Plantations in the Southern Appalachians

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Fraser fir (*Abies fraseri*) is an economically important species cultivated for Christmas tree production in the Southern Appalachian Mountains of the United States. Annual sales average about 100 million USD in North Carolina alone. Root rot and mortality caused by *Phytophthora cinnamomi* result in significant economic losses to the Fraser fir Christmas tree industry. In previous surveys conducted in 1972 and 1997–98 in North Carolina, the incidence of *Phytophthora* root rot was 9% and *P. cinnamomi* was the predominant species isolated from roots of Fraser fir. We hypothesized that the *Phytophthora* species composition in the Southern Appalachian Fraser fir production region has changed since the previous surveys because the industry has drastically increased importation of planting stock from outside of the region.

During 2014, a survey was conducted on Fraser fir Christmas tree plantations in the Southern Appalachians (NC, TN, and VA) to enumerate the *Phytophthora* species present. Using a weighted sampling strategy based on Christmas tree acreage in 14 counties, symptomatic Fraser fir roots were collected from trees in 103 commercial production fields. In total, six species of *Phytophthora* were isolated from 82 sites in 13 counties. *P. cinnamomi* accounted for 71% of the isolates, *P. cryptogea* for 23% and collectively, *P. citrophthora*, *P. europaea*, *P. pini* and *P. sansomeana* accounted for 6%. *P. citrophthora*, *P. europaea*, *P. pini* and *P. sansomeana* have not been identified in previously published Fraser fir surveys conducted in the region. While *P. cinnamomi* was still the predominant species isolated from infected Fraser fir roots, *P. cryptogea* appears to have become an important pathogen contributing to losses to the Christmas tree industry in the Southern Appalachian Mountains.

Response of Turkish and Trojan Fir Seedlings to *Phytophthora cinnamomi* and *P. cryptogea*

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Phytophthora root rot, primarily caused by the oomycete *Phytophthora cinnamomi* Rands, is a large problem for the Christmas tree industry in North Carolina, leading to more than \$US 6 million in losses annually. Fraser fir (*Abies fraseri* [Pursch] Poir.), one of the most desirable Christmas tree species in the United States, has no known innate resistance to this disease while some exotic fir species, such as Trojan (*A. bornmuelleriana* Mattf.) and Turkish (*A. equi-trojani* Aschers. & Sint) fir, display varying amounts of resistance.

A large *Phytophthora*-resistance screening trial was completed using 1600 seedlings from 12 Turkish and Trojan fir families with Fraser and momi fir (*A. firma* Sieb. & Zucc.) seedlings included as susceptible and resistant controls, respectively. Each family (or species) was inoculated with each of eight *Phytophthora* isolates, six *P. cinnamomi* and two *P. cryptogea*. The isolates were collected from a number of different diseased plant hosts (*Abies*, *Camellia*, and *Juniperus* spp.) within North Carolina. Plants were grown in Conetainer tubes under 55% shade with daily irrigation at a research nursery in Raleigh. Mortality was assessed as percent shoot necrosis bi-weekly for 16 weeks with a final observation the following year after bud break.

Overall, fir species resistance rankings confirmed previously reported results; momi fir was most resistant, followed by Turkish and Trojan fir with Fraser fir being most susceptible. *P. cinnamomi* isolates were generally more aggressive on all fir species than *P. cryptogea* isolates. There was a significant interaction between host fir species and *Phytophthora* isolates although the relative resistance rankings of fir species was consistent across *Phytophthora* isolates. *P. cryptogea* has recently become more prevalent in Fraser fir Christmas tree plantations in the Southern Appalachian region. The two *P. cryptogea* isolates used were originally isolated from Fraser fir and resulted in 50% and 100% mortality on Fraser fir in this study. Turkish and Trojan fir families appear to possess quantitative resistance to *Phytophthora* species common in North Carolina.

Mapping Phytophthora Root Rot Resistance in Fir Species through Genotyping by Sequencing

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The oomycete, *Phytophthora cinnamomi* Rands, causes root rot disease on a broad range of fir and pine species used as Christmas trees. One of the most valuable Christmas tree species, Fraser fir (*Abies fraseri* [Pursch] Poir.) has no innate immunity to *Phytophthora*, and *Phytophthora*-related damages in the Christmas tree industry add up to more than \$US 6 million annually. However an exotic fir species, Trojan (*A. bornmuelleriana* Mattf.) fir has previously shown varying amounts of resistance to *Phytophthora* root rot.

DNA was extracted from foliage of progeny in an open pollinated Trojan fir family (n=161), which was then screened for root rot resistance against *P. cinnamomi* with an overall mortality of 71%. Libraries were prepared for Genotyping by Sequencing (GBS) to identify genetic marker loci. A small subset of individuals from other inoculated Turkish, Trojan, Fraser, and momi fir families were also genotyped for comparison. The DNA libraries were sequenced on 2 Illumina HiSeq lanes, returning 342 million reads. The resulting sequence was filtered to 413,000 unique tags via the Tassel pipeline, 117,000 of which segregate within the selected Trojan fir family. The segregating tags were tested for association with the disease resistance phenotype, and significance was determined by a permutation test. 205 tags were identified as significantly associated with root rot resistance. The tags were mapped to a draft genome

assembly of loblolly pine (*Pinus taeda*), to help identify putative markers, and significant tags were also blasted against the NCBI database to identify genes with known function.

Using the 205 significant genetic markers associated with resistance, we hope to identify the genetic basis of the disease resistant phenotype. The markers associated with disease resistance in the large Trojan fir family can then be compared to the markers identified in the other fir species and families to look for consistent association of specific markers with disease resistance. The understanding of the genetic basis of *Phytophthora* root rot resistance obtained from this study will guide future breeding efforts to develop resistant planting stock suitable for use on *Phytophthora*-infested land.

Screening *Abies* for Resistance to *Phytophthora* Root Rot

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Phytophthora root rot (PRR) causes significant losses in bare-root conifer nurseries and Christmas tree plantations. True fir trees (*Abies*) are common hosts of *Phytophthora*, and popular Christmas tree species such as noble fir (*A. procera*) and Fraser fir (*A. fraseri*) are particularly susceptible. A complex of *Phytophthora* species are collectively recognized as causal agents of PRR, and vary regionally among U.S. production regions. There are limited methods available to growers for controlling PRR, but efficacy varies depending on geographic region, host species, field topography, and prior land uses. For these reasons, efforts to identify fir trees that display resistance to PRR under variable environmental conditions are justified to alleviate losses.

A large-scale greenhouse resistance screening study challenged one-year-old seedlings of 7 species of fir with 3 virulent genotypes from each of 4 species of *Phytophthora*. The *Phytophthora* isolates employed in the study were collected from fir roots during a nationwide sampling effort of tree plantations in 5 major U.S. Christmas tree production regions. In order to adequately test host performance over a range of environmental conditions, the experiment was conducted simultaneously in two greenhouses set to two different temperatures. The cool weather greenhouse was maintained at a temperature range of 15 – 21°C to replicate prevailing conditions in temperate regions such as the Pacific Northwest (PNW). The warm weather greenhouse was sustained in the 26 – 32°C range to simulate the southeastern U.S. and California. Although species such as Nordmann fir (*A. nordmanniana*) and Turkish fir (*A. bornmuelleriana*) are traditionally considered to be more tolerant to PRR than the highly susceptible noble and Fraser firs

in the PNW, evidence has shown that these species are apt to fail in other growing regions with different environmental conditions and *Phytophthora* communities. The design of this study intended to address these anomalies.

Plant material was randomized into each greenhouse in a split-split block design and inoculated by inserting colonized rice grains into the growing media. Mortality was rated weekly, and at 13 weeks all surviving seedlings were re-inoculated in the same manner. The experiment is expected to continue for an additional 5 weeks; at which time, root rot ratings and moisture content calculations should provide insight as to which seedlings qualify to be considered on the spectrum of resistance.

Tissue from resistant trees will yield genetic material appropriate for genomics testing in pursuit of molecular markers associated with resistance. It is also intended that individual trees will be conserved for future breeding applications. The goals of this study are to supplement established knowledge regarding *Phytophthora* species virulence and *Abies* sensitivities, and to enhance crop productivity by providing growers with resistant planting stock.

Fungicide resistant *Botrytis* strains are present in forest nurseries in Norway

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Fungal diseases are among the main challenges in the seedling production of Norway spruce (*Picea abies*) in forest nurseries in Norway, and grey mould caused by one or several *Botrytis* species is considered the most problematic. Control is mainly based on use of fungicides containing the active ingredient (a.i.) thiophanate methyl, fenhexamid or iprodione. A project was started in 2014 aiming to improve control of fungal diseases on spruce in forest nurseries. Due to the use of a.i.'s to which resistance has been reported in *Botrytis* strains in a number of horticultural crops in many countries, including Norway, we are currently investigating the presence of resistant strains in forest nurseries. In an initial experiment, we examined 18 *Botrytis* isolates for resistance to fenhexamid by measuring radial growth on artificial medium amended with discriminatory dosages of fenhexamid. Five isolates (18%) showed high resistance. In a second experiment, 17 isolates were examined for resistance towards fenhexamid, fludioxonil, iprodione, pyrimethanil, and thiophanate methyl by measuring germ tube growth on artificial media containing discriminatory dosages of the a.i.'s. Seven out of 17 isolates (41%) were resistant to thiophanate methyl, while the remaining ten showed moderate resistance. Four isolates were resistant to fenhexamid (24%), while two were resistant to

iprodione (12%). It was alarming that four of the isolates (24%) were resistant to two active ingredients, i.e. had developed multi drug resistance (MDR). Two MDR isolates were resistant to fenhexamid and thiophanate methyl, while the other two were resistant to fenhexamid and iprodione. Moreover, these MDR isolates also showed moderate resistance to one, two or three of the other a.i.'s. Thus far, isolates from seven nurseries have been examined, of which we found MDR in two nurseries, strains resistant to one a.i. in three nurseries, and moderate resistance towards at least one of the a.i.'s in all but one nursery (isolates from the latter were only included in the first experiment). Our preliminary conclusion is that use of thiophanate methyl should be avoided in the future, and resistance development to fenhexamid should be carefully monitored.

Identification and pathogenicity of *Phomopsis* isolates associated with spruce decline in Christmas and landscape tree settings in Michigan, USA

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Michigan is a major nursery producing state, with spruce (*Picea* spp.) being important species for both the landscape and Christmas tree industries. In the early 2000s, mature spruce in Michigan began to show combined symptoms of needle loss and branch dieback, which we term spruce decline. These symptoms had been reported on nursery and landscape spruce in Wisconsin as curling and necrosis of tips with stem cankers and on nursery and tree farm settings in Michigan as cankers but never before on mature spruce in Michigan. While most conspicuous on Colorado blue spruce (*Picea pungens*), spruce decline symptoms were also found on white (*P. glauca*) and Norway (*P. abies*) spruce throughout Michigan, as well as other states. Cankers were not associated with indentations or resinous exudates typical of canker diseases like *Cytospora kunzei*. Only when the bark layer was removed were numerous brown cankers with occasional resinous streaking seen in the phloem and cambium. Isolates from over 100 cankers on symptomatic spruce trees across Michigan were used to identify the pathogen. ITS1 and ITS4 sequencing revealed one or more *Phomopsis* species

that formed five groups differing across 11 different base-pair positions. Pathogenicity tests determined that Colorado blue spruce was the most susceptible, followed by white, then Norway spruce. Additionally, not all *Phomopsis* groups were equally virulent. Isolates from groups 2, 4 & 5 were most virulent on Colorado blue spruce, moderately virulent on white spruce and had lower virulence on Norway spruce. In contrast, the two isolates from group 3 had very low virulence levels for all spruce species tested. Group 1 showed moderate virulence on Colorado blue spruce and very low virulence levels for white and Norway spruce. Since *Phomopsis* isolates varied genetically and in virulence, future research will focus on using multiple genes to clarify the taxonomy of *Phomopsis* involved in spruce decline. As with other tree declines, we recognize that many other factors like needlecast diseases and insects could also play important roles in disease initiation and/or progression. To our knowledge, this is the first report of *Phomopsis* causing cankers associated with spruce decline on mature landscape spruce in Michigan.

Neonectria neomacrospora has caused severe damage on true fir (*Abies* spp.) in Denmark

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In 2011, typical red fruiting bodies from a *Neonectria* sp. were found on subalpine fir (*Abies lasiocarpa*) in a provenance trial in Denmark. Isolates obtained were identical to Norwegian isolated from white fir (*A. concolor*). Later the isolates were identified to *Neonectria neomacrospora* by ITS sequencing of the rDNA. The fungus was first described by Wollenweber in 1931 under the name *Nectria cucurbitula* (TODE) Fr. v. *macrospora* Wt. n. v. It has been present in Norway and North America for decades, and the imperfect stage was reported on fir elsewhere in Europe, but the detection on subalpine fir was the first time the fungus was found in Denmark. Typical symptoms and signs were flagging (dead branches) and heavy resin flow. Red fruiting bodies (perithecia) were found on several

diseased trees. No perithecia were seen on current year dieback or branches that obviously had been dead for a longer period. Perithecia were only present on branches that had died the previous year (brown needles still attached), and especial abundant where dead needles had accumulated on lateral branches. This was likely due to preservation of humidity after rain- and dewfall, creating ideal conditions for fungal growth. Koch's postulates were fulfilled on subalpine fir seedlings. Since the first finding in Denmark in 2011, the fungus has caused an epidemic and great losses on many fir species in Danish Christmas tree fields, forest stands, seed orchards and ornamental plantings. It has also been found to be seed borne and occurring in nurseries.

Development and application of a PCR-based test for the identification of *Neonectria neomacrospora* damaging *Abies* species

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In Norway, true firs (*Abies* spp.) are very important for the Christmas tree production, especially Nordmann fir (*A. nordmanniana*) and subalpine fir (*A. lasiocarpa*). In June 2008, a severe disease outbreak was discovered on white fir (*A. concolor*) in several counties in southern Norway, and identical symptoms were observed on white fir in areas of Sweden bordering southeastern Norway. A *Neonectria* sp. was isolated from the diseased trees, and sequencing of the internal transcribed regions (ITS) of ribosomal DNA showed that all isolates were identical and most similar to *N. ditissima*, a damaging pathogen in fruit orchards. The cultures obtained showed up to 99% similarity to *N. ditissima* in the ITS region, and as low as 96% similarity to *N. fuckeliana*, which has been known for decades on Norway spruce (*Picea abies*) in the Nordic countries. By the end of 2012, some new reports to the GenBank matched the sequences from our isolates from true fir, revealing that the causal organism of the epidemic on fir was *N. neomacrospora*.

In the last few years, *N. neomacrospora* has been detected and identified on many new fir species; in total 19 species and subspecies of *Abies* (Talgø & Thomsen 2015). In addition, *N. neomacrospora* has been isolated in a single case from Norway spruce (*Picea abies*) and recently also from western hemlock (*Tsuga heterophylla*) (Talgø & Brurberg, 2015).

Due to the increasing problems with *N. neomacrospora*, we have developed a Taqman real-time PCR assay specific for this fungus, for rapid identification and detection. The real-time PCR assay was optimised with various concentrations of primers and probe. The optimal concentrations gave standard curves with high correlation coefficient, indicating a reproducible linear response in detection of increasing concentrations of *N. neomacrospora* DNA. The assay was validated for specificity to *N. neomacrospora* by testing several isolates of *N. neomacrospora*, *N. ditissima* and *N. fuckeliana*. The latter two gave none or very weak signals. The assay was also tested on symptomatic plant samples from the forest and spore catches from a branch with a canker wound of sporulating *N. neomacrospora*. The assay successfully detected airborne spores of *N. neomacrospora* as well as the fungus in plant samples, and hence will be a valuable tool for identification, detection and epidemiological studies of the pathogen.

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Inoculation experiments with *Neonectria neomacrospora* on *Abies nordmanniana*

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Nordmann fir (*Abies nordmanniana*) is a widely used Christmas tree species in northern Europe. Since 2009, the bark parasite *Neonectria neomacrospora* has caused a canker epidemic on true firs (*Abies* spp.) in Norway and Denmark, including Nordmann fir Christmas tree stands. Typical symptoms and signs are dead branches, canker wounds with dead tissue below the bark, heavy resin flow, and the presence of fungal structures: cream coloured spore pustules with the conidial stage *Cylindrocarpon cylindroides* and red fruiting bodies (perithecia) with ascospores.

The fungus has also been found in Canada as a pathogen on balsam fir (*A. balsamea*) around 1960, and more recently on various fir species in the Pacific Northwest (US). The anamorph stage was first described from grafted *A. concolor* in a German nursery more than 100 years ago.

The susceptibility varies both within and amongst species, with subalpine fir (*A. lasiocarpa*), white fir (*A. concolor*), and Spanish fir (*A. pinsapo*) as the most susceptible both in field assessments and inoculation experiments on detached shoots.

Various methods for inoculation were tested on Nordmann fir, and the most effective was creating an entry point by removing a needle from the shoot, and placing a 0.5 mm plug from a *N. neomacrospora* culture upside down on the wound. This method was also successful when used on other *Abies* species. However, in general the infection success is so high, even on fully mature shoots, that it may obscure the variation in susceptibility, at least within a species. Inoculation with ascospores or conidia would imitate natural infection more closely. This is difficult to handle due to the need for freshly prepared spore suspensions, since mature perithecia cannot be stored over time for lab experiments, and conidia do not form readily on agar.

Various factors may influence the result of inoculation, such as shoot size, inoculum age, duration of experiment, humidity and temperature, wound area, and development stage of shoots. Further development of a reliable inoculation method which mirrors natural infection and pathogen-host interactions is needed in order to study the genetic variation in susceptibility of *Abies* spp. to *N. neomacrospora*. The intention is to identify highly resistant genotypes for use in breeding programs.

Neonectria – an update on genetic variation in tree susceptibility based on ocular field evaluations

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In Denmark, the fungus *Neonectria neomacrospora* was firstly identified in spring 2011 in a provenance trial of subalpine fir (*Abies lasiocarpa* ssp.) and a Nordmann fir (*A. nordmanniana*) grafted clonal seed orchard.

This newly identified fungus have caused severe problems in a number of species from the genus *Abies*, including Christmas tree stands, stands for timber production and seed orchards throughout Denmark, and also in the collection at Hørsholm Arboretum. During 2012 and 2013 a series of reports on damages were recorded, and the presumably virulence of the new damaging agent has caused serious concern in the Christmas tree industry. A number of field registrations have been carried out. For all evaluations the same scale have been used for describing the total tree damage; 0 to 10 - no damage (0), weak (1-3), moderate (4-5), severe damage (7-9), and dead (10).

In nordmann fir clonal seed orchards significant differences were seen between clones. Some genotype site interaction were found, with indications of a needle and shoot sucking aphid (*Adelgids dreifuscia*) having a role in increased fungus attack (based on a statistical correlation).

The Arboretum in Hørsholm comprises 33 different *Abies* species and subspecies, unevenly distributed. In total 360 individuals are scattered across the Arboretum collection, not randomized and uneven aged. However, despite the lack of statistical experimental design, this collection offers an interesting opportunity to evaluate the species susceptibility in a nearly even environment. A very large variation in the damage score for species was seen. The group of the most damaged species included white fir (*A. concolor*) and subalpine fir.

Seed-borne fungi on Christmas trees

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Christmas tree production has increased substantially in Norway during recent years. Species of fir (*Abies* spp.), mainly Nordmann fir (*A. nordmanniana*) and subalpine fir (*A. lasiocarpa*), have largely taken over from Norway spruce (*Picea abies*), and become the most important Christmas tree species. Noble fir (*A. procera*) is mainly grown for bough production. The presence of fungi on fir seed lots have been investigated in Norway during the last 10 years. In 2005, samples from twelve seed lots originating from Norway (three noble fir, two subalpine fir), Georgia (three Nordmann fir), Canada (two subalpine fir), Austria (one Nordmann fir) and Russia (one Nordmann fir) were tested using agar plate methods (PDA and WA). Some fungi were identified to species based on sequencing of ITS regions of rDNA. The most important finding was that *Sydowia polyspora* was present on seed from all countries; ten samples were infected, in frequencies of 0.5–87% infected seeds. This fungus is associated with current season needle necrosis (CSNN) and *Sclerophoma* shoot dieback, both diseases commonly observed in forest nurseries and Christmas tree plantations, especially on Nordmann fir. Previously, this fungus had only been reported to be seed borne on Scots pine (*Pinus sylvestris*) in Britain. *Sirococcus conigenus*, causing shoot blight of several conifer species, was found in one Norwegian *A. procera* seed lot (31% infected seeds). *Caloscypha fulgens*, the seed or cold fungus, was detected at low levels on subalpine fir seed from Canada. In addition the following fungal genera were recorded: *Acremoniella*, *Acremonium*, *Alternaria*, *Aspergillus*, *Botrytis*, *Cephalosporium*, *Chaetomium*, *Cladosporium*, *Dictyopolyschema*, *Epicoccum*, *Fusarium*, *Genicularia*,

Mucor, *Neonectria*, *Penicillium*, *Phoma*, *Rhizopus*, *Sordaria*, *Trichoderma* and *Trichothecium*. Species within some of these fungal genera are known pathogens in nurseries and production fields. In 2009, *S. polyspora* was detected in samples of pine and Norway spruce seedlings during germination tests at the Norwegian Forest Seed Center, indicating that *S. polyspora* also was seed borne on spruce. In a seed test to investigate how widespread it might be on conifer seeds, we detected *S. polyspora* in 23 out of 44 seed lots tested, representing different species within seven out of eight genera tested; *Abies*, *Larix*, *Picea*, *Pinus*, *Pseudotsuga*, *Thuja* and *Tsuga* (not on *Chamaecyparis*). In an inoculation experiment we found that *S. polyspora* may strongly reduce the emergence of noble fir seeds. In a seed treatment experiment to control *S. polyspora* on conifer seeds, using samples from two naturally infected seed lots (*Pinus mugo* var *rotunda* and *A. procera*), a fungicide containing boskalid+pyraclostrobin (Signum) was effective against the fungus without influencing the germination capacity. Testing of seeds from diseased Nordmann fir and sub-alpine fir has revealed infection by *Neonectria neomacrospora*, sometimes in rather high frequencies. This fungus can cause dead shoots, dead branches, canker wounds with heavy resin flow, and in severe cases death of trees. Serious damage has been observed in Christmas tree fields, and forest stands and seed orchards. To reduce the damages in nurseries and production fields, and to limit the risk of long distance spread of important pathogens via seed and transplant trade, surveys of seed plantations and seed health testing is recommended.

Does the severity of current season needle necrosis decrease on older stands of noble fir?

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Current season needle necrosis (CSNN) is a poorly understood disease on true firs (*Abies* spp.) grown for Christmas trees in Europe and North America. Early research suggested that CSNN was likely a physiological disorder that was associated with calcium deficiency and environmental stress. However, recent research in Norway has found that the endophyte, *Sydowia polyspora* may play a role in the development of this disease. In the U.S. Pacific Northwest (Oregon, Washington, and Idaho) and British Columbia, CSNN is most commonly seen on noble (*A. procera* Rehd.) and grand fir [*A. grandis* (Douglas ex D. Don) Lindl.] grown at low elevation sites. Similar needle damage has also been observed on white fir [*A. concolor* (Gord. & Glend.) Lindl. ex Hildebr.], Nordmann fir [*A. nordmanniana* (Steven) Spach] and Turkish fir (*A. bornmuelleriana* Mattf.). In Europe, CSNN has also been observed on grand and noble firs, but the greatest economic impact has been on Nordmann fir, the dominant Christmas tree species in Denmark and Norway.

Since 2004, the yearly severity of CSNN has been evaluated on noble fir trees in a series of genetic trials planted at WSU-Puyallup. This is a low elevation (10 to 30m) site that is very conducive to the development of CSNN and has provided an opportunity to examine yearly variation in development of CSNN and determine the variation in resistance to this disease among the different sources of trees in these trials. Unlike most trials where data was only collected for a few years, data were collected over an 8-year and 10-year period for trials that were established in 2002 and 2004, respectively. These 2002 and 2004 replicated plantings contained 25 trees from 35 and 53 sources, respectively. Starting 2 years after planting, the severity of CSNN on each tree was rated annually on a scale of 0 to 10, where 0 = no CSNN, 1 = 1-10%, 2 = 11-20%, 3 = 21-30%, ..., and 10 = 91-100% of the current season foliage damaged by CSNN.

Data from these longer-term evaluations indicated that there was significant year-to-year variation in CSNN in both plantings. There was a trend of reduced damage as trees aged in the 2002 trial and there was a significant negative correlation between the age of trees and the severity of CSNN in the 2004 trial. While the reasons for this decrease are unclear, its implications for our understanding of this disease will be discussed.

SESSION 4: PHYSIOLOGY

Does temperature at the developing bud regulate cone production in *Abies fraseri*?

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Many conifers are mast-seeding species, with seed production highly variable across years, but synchronized across a population. Lack of consistent cone production is a common problem in conifer seed orchards, and various cultural and chemical treatments have been used to increase cone production. Application of GA₄/7 + fertilizer + girdling resulted in increased cone production in Pacific silver fir (*Abies amabilis*). Cone production was further enhanced by the construction of polyethylene tents around the trees (treatment = GA₄/7 + fertilizer + girdling + tenting), suggesting that the increased temperature from solar heating played a role in cone enhancement (Owens *et al.* 2001).

Whereas seed orchard managers want to increase cone production, Michigan Christmas tree growers are looking for solutions that reduce coning. In Michigan, plantation-grown Fraser fir (*Abies fraseri*) trees produce abundant, intermittent cone crops, often beginning at an early age. Developing cones consume photosynthates at the expense of vegetative growth, and leave behind unsightly stalks and resinous scales when they disintegrate in early fall. Cones must be removed by hand, which is expensive.

In 2014, we established an evaporative cooling study at the Southwest Michigan Research and Extension Center (Benton Harbor, Michigan) to test whether the temperature at the lateral bud locally regulates cone initiation. Twenty-one randomly selected trees were misted for evaporative cooling, 12 trees were partially covered in polyethylene tents for solar heating, and 21 trees were left untreated as controls. Evaporative cooling reduced the midday temperature of misted shoots by up to 7°C. Solar heating of plastic-tented trees increased midday temperatures by up to 10°C. Effects on cone production and tree health will be presented. This research has important implications for both seed orchard managers and Christmas tree growers.

Reference

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Variation in Survival and Bud Break of Turkish and Trojan fir in the United States

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Turkish (*Abies bornmulleriana* Mattf.) and Trojan [*A. equi-trojani* (Aschers. & Sint. ex Boiss) Mattf.] fir are endemic species to Turkey. Both species have become increasingly used as Christmas trees in Europe and North America in part due to their resistance to some common diseases and insect pests. Also, both species are better adapted to warm and dry climates than native fir species and have been accepted by Christmas tree consumers. In 2010, the Collaborative Fir Germplasm Evaluation (CoFirGE) Project was organized as a partnership of university research and extension faculty and Christmas tree grower associations in five production regions of the United States (Connecticut, Michigan, North Carolina, Pennsylvania and the Pacific Northwest) and Denmark with the goal of evaluating both species for use in the Christmas tree industry. Cones were collected from three Turkish and two Trojan fir provenances in the

fall of 2010. The seeds were germinated and seedlings were grown in a greenhouse in Oregon and in 2013, two-year old seedlings were planted into two field trials in each of five production regions in the United States. The field trials are being cultured according to regional Christmas tree practices. Provenance and family variation in first year survival and bud break will be presented.

Using Herbicides to Interrupt Cone Development on Fraser Fir

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When subjected to heat and moisture stress, Fraser fir can produce heavy cone crops on immature Christmas trees. For some North Carolina Fraser fir growers, hand-pulling cones is their most labor-intensive and expensive cultural practice on a per-tree basis. There is a treatment window in spring after cones break bud and before foliage emerges when it is possible to spray cones without damaging new growth. Starting in 2012, treatments have been made during this window using different herbicidal materials that have the potential to kill immature cones. By killing cones early in their development, foliage grows more normally and hand-pulling is unnecessary. 2012 treatments included herbicides and agricultural adjuvants that had a history of damaging immature foliage without harming mature foliage. Household cleaning products that are used as herbicides by some organic farmers were also tested. Materials were applied to the tops of trees with cones using a solo backpack sprayer with an extended wand. Goal 2xl (oxyfluorfen) provided the first limited success in killing emerging cones without damaging mature foliage. Additional conventional herbicides were tested in 2013 and 2014. Labeled organic herbicides and tobacco sucker control products that burn foliage on contact were added to the treatment list in 2014.

In 2015, six of the most promising products were used: five organic herbicides and a tobacco sucker control product. Treatments included several rates of Scythe (fatty acid), Off Shoot-T (fatty alcohol), Axxe (ammoniated pelargonic salts), Avenger (citrus oil), WeedZap (clove and cinnamon oil), and industrial 20% vinegar. Seventeen treatments were applied to trees with cones at three different farms. High rates of Axxe, Scythe, Avenger, and Off Shoot-T killed more than 60% of cones at one location. When spray results at one farm were adjusted for windy conditions by omitting values for missed cones (those exhibiting no sign of any spray damage), Axxe and Scythe treatments killed more than 90% of treated cones and Avenger and WeedZap treatments killed more than 70% of treated cones. Extensive foliage injury was associated with 20% vinegar. Light foliage injury was associated with the highest rate (10% solution) of Scythe on some trees. For these potential cone treatments to be adopted by growers, they will need to work when applied by mechanized sprayers. In 2016 the best products will be tested using backpack, mistblower, and hydraulic sprayers.

SESSION 5: GROWTH CONDITIONS AND INTEGRATED PEST MANAGEMENT

Evaluating nitrogen source and application timing for optimal nitrogen uptake

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Application of commercial fertilizer materials has become an essential part of plantation management for many Christmas tree producers in Michigan. Most nitrogen applications are surface applied in the form of urea or ammonium sulfate. With surface applications of nitrogen there is potential for nitrogen loss from volatilization, leaching and denitrification. Some growers have begun to add nitrogen stabilizers due to concerns over the loss of nitrogen, hoping to reduce nitrogen loss and optimize plant uptake. The addition of nitrogen stabilizers can add an additional \$70.00+ per ton. We established trials in the fall of 2013 to determine if timing of nitrogen application or the choice of nitrogen fertilizer products influ-

enced growth or foliar nitrogen values. The nitrogen fertilizer sources that were applied were urea, stabilized nitrogen (SuperU®) and ammonium sulfate. The stabilized nitrogen source is designed to slow nitrate-N loss by including urease and nitrification inhibitors. These products were applied as split applications fall/spring or just as a spring application. In the fall of 2015 leader and lateral length and foliage samples were collected. Our initial results indicate that shoot growth did not differ among any of the treatments, including the unfertilized control. Fertilization increased foliar nitrogen levels compared to unfertilized controls but there was no difference in foliar nitrogen among fertilizer treatments.

Integrated Pest Management Education for the Christmas Tree Industry in Oregon, USA

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The Pacific Northwest is the country's largest Christmas tree producer. Oregon has 63,000 acres of Christmas trees, with a sales value of \$110 million in 2013. In addition to the ongoing applied research, education for workforce and grower awareness about export threats are priority areas for Extension and outreach at the NWREC. Two new bilingual (English and Spanish) publications have been produced in the last three years to support these educational objectives. In 2012, with grant support, the field guide *Identifying and Managing Christmas Tree Diseases, Pests, and Other Problems*, was developed to help Christmas tree growers and field workers implement pest management activities. It features descriptions of diseases, pests, disorders, and damage affecting Christmas trees in the Pacific Northwest, and describes how to identify and manage these problems. It includes management calendars, susceptibility scales, over 100 color photos, and a glossary of terms. It was designed in a pocket-size, flip book format on waterproof paper for field use. Three hundred copies were produced and sold out the following year. This first edition was revised and published by Extension & Experiment Station Communications (EESC) at Oregon State University in April 2014 (PNW659). It was accepted as a Pacific Northwest Publication (Idaho, Oregon and Washington) because of the interest for the entire region.

A second field guide, *Best Management Practices for Christmas Tree Export*, was developed to provide information on identifying and managing pests of concern to export trading partners. It features best management practices to help minimize the presence of pests at harvest and describes how to identify these problems. It includes management calendars, pest quarantine information, legal considerations for exporting, and options for monitoring and trapping. It was also published by EESC in July 2014 (EM 9093). These two publications won the 2015 Silver Award in the diversity category from the Association for Communication Excellence, an organization that recognizes professional work and service in agriculture.

These two guides are intended to facilitate better communication between English and Spanish speakers and help ensure successful harvests. They have been an excellent resource to support regional IPM trainings for the Christmas tree industry, particularly targeting crew leaders and workers that only speak Spanish. Topics covered in these trainings included field scouting, pest identification, best management practices for export, use of digital microscopes, and sampling protocols. Follow-up evaluations found a 75% improvement in worker knowledge associated with scouting of Christmas tree problems after trainings.

Management of diseases in Norwegian Christmas tree fields

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To control disease problems in Christmas trees, good knowledge about culturing methods, plant material (e.g. species and provenances), climatical conditions, soil- and nutrient parameters, biological and chemical control methods etc. is required. The best management practice is often a holistic, environmental friendly approach or so-called integrated pest management (IPM). In IPM, the goal is not to eradicate all damaging agents, but to keep the impact below a threshold level. First, healthy transplants are of vital importance to give the production a good start and to avoid introduction of new diseases that may follow the nursery stock. Furthermore, correct identification of disease causing agents is crucial for implementing proper management. Management of air- and soil borne diseases requires different approaches. In general, airborne diseases may be kept at a low level by decreasing air humidity in and around the trees by planting parallel to the most predominant wind direction and not too dense. Good weed control and pruning of lower branches (making of handles) will also add to more rapid drying of foliage after precipitation. Some herbaceous weed and broad leaved trees serve as alternate hosts to rust fungi, e.g. rosebay willow herb (*Chamerion angustifolium*) and *Epilobium* spp. for silver fir needle rust (*Pucciniastrum epilobii*), bird cherry (*Prunus padus*) for cone rust (*Thekopsora areolate*) and willow (*Salix caprea*) for fir-willow rust (*Melampsora abieti-capraearum*). The rusts may be managed by controlling the alternate hosts, mechanically or by herbicides, inside and in the vicinity of Christmas tree fields. To minimize the use of pesticides (herbicides, fungicides and insecticides), application of fungicides and insecticides to individual trees should be considered, especially if the problem is not yet widespread. To avoid problems

getting out of hand, regular monitoring for potential damaging agents is vital. The most vulnerable stage concerning both biotic and abiotic damages is during shoot elongation. That is the period when fungicide application may be necessary to control fungi like *Delphinella* shoot blight (*Delphinella abietis*) and *Neonectria* canker (*Neonectria neomacrospora*), especially in humid, coastal regions. In general, to keep the disease pressure low, pruning of diseased shoots, branches and in severe cases removal of whole trees is advisable. Root and soil borne problems are mainly due to the diseases *Armillaria* root rot (*Armillaria* spp.), annosus root rot (*Heterobasidion annosum*) and *Phytophthora* root rot (*Phytophthora* spp.). They can all do considerable harm, but due to survival in soil for decades even without the preferred host, *Phytophthora* root rot is considered the most devastating of the three pathogens. For the former two diseases, stump removal before replanting is a good management strategy. Against *Phytophthora* root rot, well drained soil may reduce the impact, but in wet areas on heavy soil *Phytophthora* spp. will have enough moisture to thrive however well the soil is drained. Selection for more resistant hostplants is probably the best approach for the future.

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SESSION 6: POSTHARVEST, MARKET AND ECONOMY

Postharvest moisture status and quality of trees displayed in tenon-type Christmas tree stands

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Displaying Christmas trees in water holding stands has been shown to be an effective way of maintaining tree freshness, minimizing needle loss and reducing fire hazards associated with displayed trees. Water uptake during display is influenced by a number of factors, including tree species, the moisture content of the tree when it is set up, the temperature and relative humidity of the display area, how long it has been since the base of the tree was cut, the water-holding capacity of the stand, and the care the tree receives during display.

During the past few years, there has been an increased use of tenon-types of water-holding stands to display table-top trees in the United States. These stands have been used in Europe for a number of years and the concept behind them is to use a commonly available cutter to shave the end of the stem down to a uniform sized tenon that varies in length and diameter depending on the cutter that is used. The tenon is then inserted into a receptacle in the stand. In the U.S., table-top trees are sold already attached to the stand. Consumers select a tree, take it home and add water to the stand.

During the past two years, we have conducted postharvest display trials with noble (*Abies procera* Rehd.), Fraser [*A. fraseri* (Pursh) Poir], and Nordmann fir [*A. nordmanniana* (Steven) Spach] table-top trees to determine what effect tenon stands have on their freshness and quality. Trees with freshly-cut bases that were displayed directly in water maintained high moisture level and quality ratings throughout the 10 to 14 day trials. However, trees that were displayed in the water-filled tenon stands had similar moisture levels and quality ratings to trees that were displayed without water. These trees dried rapidly and by 7 to 10 days, they had dried to the point that they posed a fire hazard. Results from these trials indicated that displaying trees in water-holding, tenon-type stands was a very ineffective way of maintaining the freshness and quality of displayed trees.

Inferring gene networks regulating needle abscission in Fraser fir through RNA-seq data

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Needle abscission (NA) is a plant physiological process that involves a few layers of cells in specific sites called abscission zones (AZ) where needles are shed. This process is triggered by endogenous factors combined with a variety of environmental signals and stresses. Fraser fir (*Abies fraseri*) has been ranked as one of the most popular Christmas tree species sold in North America and represents over 90% of all the trees grown in North Carolina as Christmas trees. Although postharvest needle retention has a prominent role in the Christmas tree industry's competitiveness with artificial trees, the physiology of the process is currently vaguely defined, and the underlying control mechanisms and gene regulatory networks are completely unclear. Fir species vary considerably in needle holding ability. Needle loss data show a high level of tree-to-tree variation among populations and individuals, and a high correlation from year-to-year within individual, suggesting a strong genetic component of this phenotypic variation. Next-generation sequencing (NGS) technologies are defining new breeding strategies for plants and animals. NGS of RNAs (RNA-Seq) is a powerful approach to determine the relationship between the coded information in a genome, its expression and phenotypic variation. To identify key regulatory genes, we constructed cDNA libraries using AZs from Fraser fir trees that exhibit

good and poor needle retention based on three years of previous phenotypic data. Differentially expressed genes that characterized trees with good or poor needle retention will be clustered into functional groups and used to reconstruct novel gene regulatory networks controlling needle abscission in Fraser fir. We are currently testing new cluster approaches and including data from other firs that exhibit extreme needle loss. Our main goal is predict the needle retention behavior of a tree based on the expression of a pool of genes. Once a proven genetic marker system has been developed, a modeling analysis will be carried out to compare the cost and time savings of the new system relative to current assessment methods.

Seasonal changes in balsam fir needle abscission patterns and links to environmental factors

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Several studies have suggested that postharvest needle retention increases in autumn, likely due to cold acclimation. But some of the dynamics of the phenomenon have yet to be studied. The objectives of this study were to (1) describe seasonal changes in postharvest abscission, water uptake, fluorescence, and moisture content of balsam fir, (2) determine the relationship of needle abscission with water uptake, fluorescence, and moisture content, and (3) link the postharvest changes to certain environmental factors. Branches were collected from 18 trees each month and needle abscission, fluorescence (fv/fm), water uptake, and water content were monitored for 12 weeks. Fluorescence, water uptake, and water content were all correlated with needle abscission throughout the study, which added to their value as strong indicators of postharvest quality.

Further, the above 4 factors were all improved in autumn months compared to spring or summer months and strongly related to changes in photoperiod. It is suggested that photoperiod has the most influence on seasonal changes in postharvest quality, with only little improvement attributed to temperature. Average needle abscission commencement can be described as a function of photoperiod using $y = 109.3 - 5.7x$ ($R^2 = 78\%$) and average needle retention duration can be described as a function of photoperiod using $y = 108.3 - 4.9x$ ($R^2 = 92\%$). Since each function describes a strong, negative, linear relationship it can be suggested that the shorter the photoperiod during the harvest of Nova Scotia grown balsam fir, then longer it will take for postharvest abscission to commence and complete. This implies that the ideal date for harvest would be December 21st and superior needle retention would be found in trees harvested close to this date.

The European Christmas tree industry – aspects of markets and production

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The European Christmas tree industry is organized in Christmas Tree Grower Council of Europe (CTGCE) which was founded in 1989 for individual members but in 2002 was changed to an organization for national associations. Today 13 countries are members of CTGCE – see www.ctgce.com for further information.

The statistics for Christmas tree production in Europe are difficult to estimate since no Europe-wide report system is in place. Therefore, overall figures for the production are based upon national reports and these figures can be variable due to Christmas trees being either forest or agriculture.

We estimate that Christmas trees are produced on some 115,000 ha in Europe resulting in an annual production of approximately 75 million Christmas trees. The production is mainly focused on fir, with Nordmann fir as the most dominant of these. Among the spruces, Norway spruce and Blue spruce are most commonly used. Germany has the largest production of Christmas trees in Europe whereas Denmark is the second largest and the biggest exporting country for Christmas trees in Europe.

Production cost for growing Christmas trees is highly dependent on labor costs. However even with high Danish labor costs it can be feasible to grow Christmas trees in comparison with many agricultural crops.

The challenges of establishing a niche market for CHO.C.O. (CHOose, Cut, Offset) Christmas trees farms in Greece

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Greece used to have a small- but quite significant for some of its semi-mountainous areas -real Christmas trees market. The producers of these cultivated trees (mostly fir, *Abies borissi-regis*) achieved a good income by selling their product in Athens and Thessaloniki, and the consumers were satisfied that they had the chance to have a real Greek tree for the Christmas time. However, due to several reasons such as the lack of suitable marketing, competition with artificial trees, financial crisis, producers getting older, low tree quality and high price, there is a significant decay in the market, which every year becomes worse and worse. On the other hand, research on real Christmas trees in Greece, if any, has been very limited, and has not given any alternatives to the producers, or the consumers. This year (2015) is the first time that there is a fund for conducting a research in the Forest Research Institute of Thessaloniki on how to offer a new product for both the supply and demand side. The aim is to produce guidelines on how to implement the Choose & Cut farms for real Christmas trees in Greece, a product which is very popular in the United States of America, and Canada; how to enhance this experience by offering the op-

portunity to producers and consumers to offset the negative environmental impact that is produced by the activity; and how to certify that the whole activity is carbon neutral. The main objective of the research project CHO.C.O. (CHOose, Cut, Offset) is to establish a niche market where some of the producers who are located close to urban areas will use a part of their Christmas tree cultivation to offer the experience of CHO.C.O. farms to a part of consumers who want to live such an experience, instead of just buying their tree in the city. The research program lasts until November 2015 and among its deliverables there is the production of a handbook for the producers and for the Public Forest Service, which supervise the harvest and transportation of the real Christmas trees, and the dissemination of the research, mainly to the producers. In this paper we will present the preliminary results of the research program, and we will try to give some guidelines for the adoption of the CHO.C.O. farms by the more experienced in real Christmas trees countries.

“Fjordtree”

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The Norwegian Christmas tree grower association applied in 2012, together with the two biggest wholesalers, for grants to finance the “Fjordtree”-project. The objective of the project was to build “Fjordtree” as a brand for Norwegian grown subalpine fir (*Abies lasiocarpa*) in the premium segment of the export market. Grants and internal funding gave the project a total budget of NOK 4,500,000 (EUR 560,000).

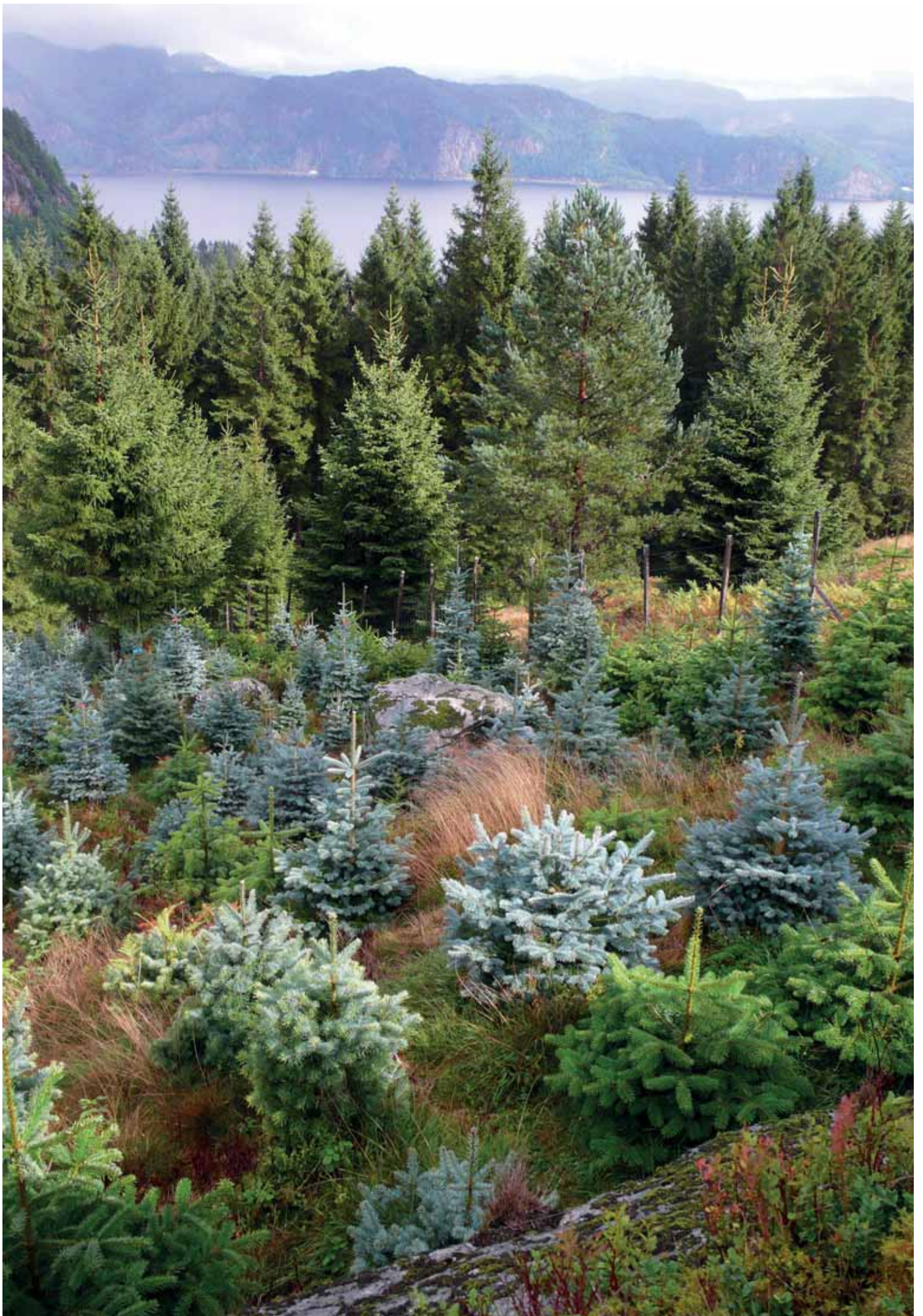
Norway spruce (*Picea abies*) has traditionally been the Christmas tree of choice in Norway. Then Nordmann fir (*A. nordmanniana*) became popular, but in 2012 it was anticipated that a new tree would dominate the market in the future. One of the candidates was subalpine fir, that already had been planted in a big scale in Norway. It was therefore a necessity to secure a market for this tree. It was also expected that the scale of the planting previous years would exceed the national market, and export would be a crucial outlet the following years.

According to the prognoses we should see the rapidly growing supply of subalpine fir already now, but it seems to be absent. We are currently investigating if this is because the trees are coming later on the market due to earlier climatic problems and/or consecutive hard shearing, or that large portions of the plantings have been taken out of production due to other reasons.

We conducted the first export to Germany in 2014 of 1.500 trees of the Fjordtree brand. The outlet price in Germany was approximately NOK 1200 (EUR 150). The reception was extremely uplifting and it is already demand for more trees than we can deliver for 2015.

The brand is currently owned by:

- Norsk Juletre (The Norwegian Christmas tree grower association)
- Norsk Juletre Service AS (wholesaler)
- Ligos KS (wholesaler)







**ABSTRACTS
POSTER PRESENTATIONS**

Variation in postharvest needle retention characteristics of Turkish and Trojan fir populations from Turkey

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Postharvest needle retention is an important attribute of Christmas trees. Previous studies with Nordmann fir have shown that needle retention is under strong genetic control and that progeny from open-pollinated trees with superior needle retention also tend to exhibit the same characteristic. In 2010, cones and branches were collected in Turkey from three Turkish fir (*Abies bornmuelleriana* Mattf.) populations (Adapazarı-Akyazı, Bolu-Alada and Karabük-Keltepe) and two Trojan fir [*A. equi-trojani* (Aschers. & Sint. ex Boiss) Mattf.] populations (Çanakkale-Çan and Balıkesir-Kazdaı) as part of the international Collaborative Fir Germplasm Evaluation (CoFirGE) Project. Collections were made from 20 different trees, representing a range of elevations within each population, during the first week of October. As much as possible, cone-bearing trees showing good Christmas tree form and growth traits and spaced at least 100 meters from one another were selected to reduce relatedness. In addition to collecting cones and making a number of measurements on each of the mother trees at the time of cone collection, 4 branches were collected from each tree. The branches were collected from the upper third of the crown where each had good exposure to sunlight. To assess differences in needle retention among the

trees, subtending lateral branches (“tongues”) were harvested from each branch. These were transported to Ankara and displayed without water in a room that was maintained at about 20 °C. After 10 days, the branches were gently rubbed between fingers three times and the severity of needle loss for each age class of needles (2009 and 2010) was rated according to the following scale: 0 = no needle loss, 1 = < 1%, 2 = 1-5%, 3 = 6-15%, 4 = 16-33%, 5 = 34-66%, 6 = 67-90% and 7 = 91-100% needle loss.

Needle loss ratings among the individual trees from the Adapazarı-Akyazı, Bolu-Alada and Karabük-Keltepe Turkish fir populations ranged from 0 - 6.8, 0 - 6.3, and 0 - 5.6, respectively. The ratings for the Çanakkale-Çan and Balıkesir-Kazdaı Trojan fir populations ranged from 0 - 5.6 and 0 - 3.6, respectively. The percentage of trees within each population that had needle loss ratings <1 ranged from 35 to 50.5%. There was no difference between elevation and needle loss ratings among any of the populations of trees. This baseline data will be compared with future needle loss data collected from the progeny growing in U.S. and Denmark in common garden studies.

Growth and postharvest needle retention characteristics of balsam fir grown in western Washington

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In 2008, a replicated common garden field trial was established at the Washington State University Research and Extension Center in Puyallup, WA to evaluate the growth and postharvest characteristics of 26 provenances of balsam fir [*Abies balsamea* (L.) Mill.] and eight progeny collections of ‘bracted’ balsam fir [*A. balsamea* var. *phanerolepis* (L.) Mill. var. *phanerolepis* Fernald]. A single source of Fraser fir [*A. fraseri* (Pursh) Poir] was included in the trial as a standard. Seed was obtained from the Canadian Forest Service’s National Tree Seed Center (NTSC) and P+2 seedlings were out-planted in February of 2008 in a 0.44 ha plot at 1.8 m x 1.8 m spacing. The plot design was a randomized complete block with five blocks. Five trees of each source were planted in a row within each block. To obtain information on adaptability to growing conditions in western Washington, data were collected on growth, bud break growing-degree days (GDD), and color. Tree form and commercial grade were assessed in 2014, and were used to estimate the wholesale value of each tree. During fall 2012 and 2014, two branches were harvested from each tree and displayed dry to determine the postharvest needle retention characteristics of each tree. Needle loss was rated on a scale of 0 (none) to 7 (91–100% loss).

All of the balsam sources broke bud prior to Fraser fir and there was a significant differences in bud break GDD among the balsam sources. In 2014, tree heights ranged from 1.5 to 2.1 m and there was no significant difference in foliage color. Seed source had a significant effect on the estimated commercial value of trees. Average values by seed source ranged from \$14.74 to \$27.34. There was considerable variability in value within regional seed sources. Four of the five highest value seed sources and four of the five lowest value five seed sources were from New Brunswick. The average 2012 and 2014 needle loss ratings for the seed sources ranged from 1.4 to 4.0. Although trees from the NTSC No. 20021377 seed source were among the top five when rated for value, this source from Fairview, New Brunswick had the highest needle loss rating.

Even though WSU Puyallup is outside of the natural range of balsam and “bracted” balsam firs, this study indicates that there are sources of these species that are well adapted for the production of Christmas trees in western Washington. Given that a seed source with a high tree value did not always have acceptable post-harvest needle retention, care needs to be taken when selecting seed sources in order to insure the best tree quality as well as profitability.

Effectiveness of hot water dips to eliminate slugs on exported Christmas trees

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In the United States, the Pacific Northwest (PNW) region leads the nation in the production of Christmas trees. Over 90% of the trees produced are either shipped throughout the U.S. or exported to a number of foreign countries. For example, in 2012 the Oregon Department of Agriculture and Washington State Department of Agriculture issued 2,349 and 66 federal phytosanitary certificates, respectively. Trees were shipped to 17 countries, with the bulk going to: Mexico (2,243), Canada (42), Hong Kong (41), Japan (17), and Singapore (18). A total of 283 container loads were also shipped to Hawaii.

Although most exported trees are mechanically shaken prior to shipping to reduce the risk of certain “hitchhikers” such as yellowjackets and slugs, the presence of slugs on exported trees has become a major issue in Mexico and Hawaii. In addition to mechanical shaking of unbaled trees, there has been some interest in using a “hot water shower” treatment that was developed to treat potted plants that are infested by an invasive coqui frog (*Eleutherodactylus coqui*) to rid trees of slugs. In 2012, 25% of the 67 quarantined containers that were treated in Hawaii were given a “hot water shower” at 47.7°C for 8 minutes. While this hot water treatment appeared promising, the system was very labor intensive and costly.

In 2013 and 2014, a series of trials were conducted to examine the effectiveness of hot water dips in killing slugs on Christmas trees. Slugs were immersed in water that was heated to temperatures between 34.4 to 51.1°C for periods between 15 seconds and 12 minutes. Checks consisted of slugs immersed in water at 12.8°C. Noble fir (*Abies procera* Rehd.) and Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco] branches were also included in these tests to determine if the treatments had any adverse effects on the foliage. The shortest exposure duration/temperature that resulted in 100% mortality of all the slugs was 30 seconds at 47.7°C. A 2 minute exposure was required at 41.1°C to kill all of the slugs. Damage was only observed on branches that were exposed to >44.4°C for more than 2 minutes. These data indicated that short-duration hot water dips could be used to reduce the risk of spreading slugs on exported Christmas trees.

Delphinella shoot blight and Grovesiella canker on *Abies lasiocarpa* in western USA

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Delphinella shoot blight, caused by the fungus *Delphinella abietis*, attacks several species of true fir (*Abies* spp.) in North America and Europe. The fungus kills current year needles, and in severe cases entire shoots, and dead needles become covered with numerous, black pseudothecia. Grovesiella canker (*Grovesiella abieticola*) results in dead shoots and branches on fir and can eventually kill whole trees. In Europe, the fungus has only been found in Poland (Sieber & Kowalski 1993). In 2013, in a provenance trial of subalpine fir (*Abies lasiocarpa*) and corkbark fir (*A. lasiocarpa* var. *arizonica*) at Sandpoint, Idaho, disease estimates for both pathogens were carried out on a scale from 0 to 3 (0 = no damage, 1 = minor damage, 2 = medium damage, 3 = severe damage). Some trees were dead or missing. No attempt was made to identify the cause of mortality since most of them had already been dead for a long period. In previously published material from the provenance trial at Sandpoint, the disease described as a *Phoma*-type blight (Barney *et al.* 2013) was most likely *D. abietis* and *G. abieticola* was not mentioned. The seed sources originated from the Rocky Mountain states of Colorado, Utah, and New Mexico and from the mountains of Arizona. The field trial was established in 2001 with a total of 960 trees [3 replicates (blocks) of 16 randomly distributed subplots (seed sources) with 20 trees in each]. The 16 seed sources included six corkbark fir (Apache-Sitgreaves, Cibola, Coconino, Coronado, Gila and Santa Fe) and ten subalpine fir (Arapaho, Carson, Cibola, Dixie, Kaibab, Manti-La Sal, Rio Grande, San Isabel, San Juan, Uncompahgre). Corkbark fir grows in native

stands between 8000 – 12000 feet (2438 – 3658 m), while subalpine fir is found between 2000 – 11000 feet (610 – 3353 m). Significant differences between provenances in susceptibility to *D. abietis* and *G. abieticola* were observed. In general, subalpine fir was more susceptible to both diseases than corkbark fir. This was also reported by Barney *et al.* (2013) for the *Phoma*-type blight, and corresponds well with results from a provenance trial in Norway, where the general outcome was that susceptibility to *D. abietis* decreased with increasing altitude of the seed source and increased with the latitude (less blue/waxy varieties) (Talgø *et al.* 2015). The four subalpine fir provenances of Uncompahgre, Manti-La Sal, Dixie and Arapaho were more susceptible to both diseases. In 2013, *D. abietis* was also found on subalpine fir in the lowland of Washington State, but neither disease was detected in native stands at Mt. Rainier, Mt. Spokane, Sherman Pass or Frazer Creek.

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Utilizing webinars to increase the adoption of integrated pest management

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In October 2013, Michigan State University Extension launched an online, on-demand series of webinars focused on increasing grower and educator awareness of IPM (integrated pest management) resources, practices, history and implications. From December 2013–December 2014, available webinars include; Introduction to Integrated Pest Management, Integrated Pest Management Resources, Entomology 101, Plant Pathology 101, Soil Science 101, Plant Science 101 and Insect Scouting in Fruit Crops. Webinar viewing was incentivized by partnering with the Michigan Department of Agriculture and Rural Development to provide continuing education credits for certified pesticide applicators. This approach to content delivery proved popular and allowed MSU Extension to access traditionally underserved audiences in Michigan as well as new national and international

participants. The program was evaluated using an online pre- and post- survey of viewers. During the first ten months, there were 1,663 webinars viewed. An approximate 430 viewers reported an acreage impact of 1.2 million acres. Approximately 30% identified as growers, 20% landscapers, 19% recreational gardeners, 13% crop consultants, 10% agriculture educators, 8% general public, 5% pesticide distributors, 3% students, and 0.4% policy makers. Based on the preliminary evaluation of the MSU IPM Webinar Series, prerecorded and on-demand webinars offer an affordable and accessible way for stakeholders to access University resources and an efficient means for garnering a wider audience for those resources and increasing the adoption of IPM practices.

Lipid and fatty acid changes linked to postharvest needle abscission in balsam fir

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Needle loss after harvest is a major problem for Atlantic Canada's balsam fir Christmas tree and greenery industry. Lipid and fatty acids constitute membrane integrity and any change in membrane integrity is reflective of the changes in lipid and fatty acid composition triggering physiological dysfunction in cellular trafficking. It is hypothesized that lipids and fatty acid compositional changes trigger and/or modulate postharvest needle abscission in balsam fir. Balsam fir branches were collected from a clonal orchard in Debert, NS, and kept hydrated in the lab at an average temperature of 20–24°C and a light intensity of 85–95 $\mu\text{mol m}^{-2}\cdot\text{s}^{-1}$ supplied by incandescent and fluorescent lights. Parameters including needle loss and water use were measured initially and three times a week for 11 weeks. Membrane injury (MII) was measured initially and once a week. In addition, needles were sampled on site, and at the start of abscission and again at peak abscission postharvest and analyzed for polar lipids and fatty acids (FAs). Peak abscission was estimated at 11 weeks postharvest. During this time water use decreased by 67%, and MII increased

by 134%. Total polar lipids significantly decreased from 53.3 $\text{nmol mg}^{-1}\text{ DW}$ to 6.6 $\text{nmol mg}^{-1}\text{ DW}$ ($p = 0.00$). This could be due to degradation, reutilization or membrane remodeling, and/or inhibition of lipid biosynthesis. Galactolipids found primarily in the chloroplasts, monogalactosyldiacylglycerol (MGDG) and digalactosyldiacylglycerol (DGDG), showed the greatest decline; 17.6 to 0.8, and 14.7 to 0.9 nmol mg DW^{-1} , respectively, suggesting predominantly chloroplast membrane breakdown. MGDG and DGDG composed 33% and 27.5%, respectively, of total polar lipids in fresh needles. This dropped postharvest to 11.5% and 13%, respectively, at peak abscission. There was a significant decrease in α -linolenic acid ($p = 0.000$), the main FA comprising MGDG and DGDG. α -linolenic acid ($\text{nmol mg}^{-1}\text{ DW}$) correlated inversely with needle loss ($r_p = -0.910$) Unsaturated: saturated fatty acid ratios changed significantly ($p = 0.000$) from 4:1 to 1:1 from initial sampling to peak abscission. There was also an inverse correlation between the ratio of unsaturated: saturated fatty acids and needle loss ($r_p = -0.843$).

A summary of BCTGA UK trials in Christmas trees 2013 to 2015

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With many UK Nordmann Fir plantations now entering their third rotation, pests, diseases and weeds have become an increasing problem for growers. To improve the knowledge base in the UK the BCTGA have conducted a development programme to study these problems and identify potential solutions.

Initial studies have included trials on Current Season Needle Necrosis, Silver Fir Woolly Aphid *Dreyfusia nordmanniana*, improved crop establishment, glyphosate rainfastness, crop tolerance to post emergent herbicides, Nordmann Fir leader control and optimum Nordmann Fir provenance selection.

The BCTGA wishes to thank the many agrochemical companies who provided products for these trials, and to Maitland who provided the plants for the provenance studies. Particular thanks go to Agrovista, Bayer Environmental Science, Nufarm UK and Nutrel UK who also sponsored some of this work.

UK trials 2013 to 2015 for the control of Current Season Needle Necrosis (CSNN) in Nordmann Fir using fungicides and calcium treatments

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CSNN has been a minor problem in the UK for many years, but since 2011 many UK plantations have been severely affected, with particularly bad attacks in 2012 and 2013. Previous work has identified that the associated pathogen *Sydowia polyspora* can be controlled by some fungicides in the laboratory, but effectiveness of fungicides has not been confirmed in the field. UK Field trials were conducted in 2013 and 2014 using multiple applications of a wide variety of fungicides from a diverse range of fungicide groups including triazoles, strobilurins and mixtures at autumn and spring applications. The work confirmed that field applications of fungicides at these timings and rates did not appear to control the condition. However, severe crop scorch was recorded from a number of applications, particularly EC and SC formulations of triazoles including cypraconazole and tebuconazole applied at bud swelling.

In 2015, work was predicated on reducing pathogen access by using soil and foliar applications of calcium nitrate, with foliar applications applied with and without additives claimed to have been successfully used in other horticultural sectors. The results from these applications are currently being analysed.

UK trials 2013 to 2015 for the control of Silver Fir Woolly Aphid *Dreyfusia nordmanniana*

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Past control of the Silver Fir Woolly Aphid *Dreyfusia nordmanniana* in the UK has largely been through the use of synthetic pyrethroids in January / February or at full flush. These control strategies were felt to be sub optimal, and likely to reduce the populations of beneficial predatory insects. Field trials were designed to suggest effective control through better selection of insecticide and accurate timing.

In 2013, trials showed that deltamethrin in early April provided excellent control of the pest, while the neonicotinoid acetamiprid was highly effective post flushing, with less effect of predatory insects expected.

In 2014 & 2015, trials confirmed the deltamethrin effectiveness and timing, and found thiacloprid to be the superior neonicotinoid for use during flushing. The 2014 trials also assessed the effectiveness of basal sprays and soil injection of systemic insecticides, used to minimise effects on non target insects. Both techniques provided acceptable results, but tended to be less effective than foliar sprays.

The 2015 field trials also included August applications of acetamiprid and flonicamid applied to the late summer generation prior to wax formation. Results from these applications are currently being assessed.

Baiting for *Phytophthora* in waterways associated with Christmas tree production in Norway, Belgium and Denmark

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Phytophthora root rot caused by several *Phytophthora* spp. is a serious disease in the fir (*Abies* spp.) Christmas tree and bough production in USA, but there are very few reports from Europe. However, in Norway we found that *P. cambivora* killed noble fir (*A. procera*), for bough production and *P. megasperma* and a *P. inundata*-like species killed subalpine fir (*A. lasiocarpa*) and Nordmann fir (*A. nordmanniana*) Christmas trees, respectively (Talgø & Chastagner 2012). In the latter case, there were clear indications that the pathogen had arrived by imported nursery stock. In Europe, there is an extensive trade with fir seedlings, thus, *Phytophthora* spp. may also become a problem for the Christmas tree industry in other European countries. A simple way to survey for *Phytophthora* spp. in an area is by baiting in waterways. Thus, leaves from *Rhododendron* 'Cunningham's White' were used as baits in Christmas tree fields in Norway in 2012. They were placed in net bags and left in ditches, small streams or drains for one week. Each bag had a styrofoam floating device to keep the baits near the surface. After baiting, black or water soaked spots on the *Rhododendron* leaves, were taken as indications for *Phytophthora* infection. From the

leading edges of such spots several *Phytophthora* spp. were isolated; *P. plurivora*, *P. gonapodyidis*, *P. inundata*, *P. cryptogea* and some isolates that we were not able to identify to species level. In 2015, baiting associated with the Christmas tree production was carried out in Belgium and Denmark. This resulted in *P. cambivora* and *P. gonapodyides* from Belgium and *P. lacustris*, *P. gonapodyides* and *P. syringae* from Denmark. *Phytophthora* spp. may be present in an area without causing mortality to fir, especially on well-drained soil, but if infested water is used for irrigation, or floods occur, the infection level may rise. Impact also depends on how resistant the host plant is and the aggressiveness of the *Phytophthora* sp. in question.

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Scleroderris canker found on Nordmann fir in Norway

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In 2013, scleroderris canker caused by *Gremmeniella abietina* was found on Nordmann fir (*Abies nordmanniana*) in a Christmas tree field in Vest-Agder County in southern Norway. *G. abietina* has never been reported on this host in Norway or elsewhere. Pycnidia with conidial spores of the *Brunchorstia* stage of the fungus were detected around dead buds. The bark of the diseased area had resin droplets and was slightly sunken and cracked. There was a sharp margin between dead and living tissue, from where the fungus was isolated. The fungus was also isolated from spore mass. The internal transcribed spacer region (ITS) sequence of the nuclear ribosomal DNA from the Norwegian isolate from Nordmann fir was identical (100%) to several *G. abietina* reported to GenBank, but different from *G. abietina* var. *balsamea* (less than 97% similarity). *G. abietina* attacks a number of conifer hosts in Europe and North America, especially pine (*Pinus* spp.). In Norway, the latest epidemic by this fungus occurred on Scotch pine (*P. sylvestris*) in 2001 in the southeastern part of the country. The following year, Norway spruce (*Picea abies*) seedlings damaged by *G. abietina* were found in forest nurseries in the same area (Talgø & Stensvand 2003). *G. abietina* var. *balsamea* has been

reported on balsam fir (*A. balsamea*) in North America and on Sachalin fir (*A. sachalinensis*) in Japan (EPPO 2009). Infections by *G. abietina* may occur via wounds throughout the growing season, but primarily new needles become infected in June–July in Norway. From the needles, mycelia may grow into the shoots and cause canker wounds and sometimes girdling. The development of disease primarily takes place at low temperatures, and the fungus may grow down to 0°C. Thus, symptoms may not be prominent until the next spring, and therefore infected seedlings may become marketed with latent infections.

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Characterization of postharvest quality of two French Christmas Trees: Nordmann fir (*Abies nordmanniana*) and Norway spruce (*Picea abies*)

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Currently, 90% of all Christmas trees sold in France are cut trees. Consumers buy their firs earlier and earlier, but they want them still “fresh” until the beginning of January. The “freshness” notion covers criteria such as needle density, colour and brightness. The needle losses after consumer purchase is a major concern to the industry. Therefore, it is important to better understand the factors involved in maintaining the quality, upstream and downstream the sale of the Christmas Tree. The objective of this study was to characterize the “freshness” as to find criteria for warrant the quality, on the two majors species grown and sold in France: *Abies nordmanniana* and *Picea abies*. Three provenances with contrasted climate, from oceanic to continental (Finistère, Beauce and Morvan), were compared to evaluate the environment effect on trees postharvest quality. Biophysical and physiological criteria such as needle water content, xylem pressure potential, needle colour, biochemical content, were evaluated periodically throughout the harvest, distribution and consumption phases. Photosynthetic activity

was measured just before the harvest. Temperature and hygrometry were continuously monitored during the postharvest phases. Global esthetical aspect of trees was also evaluated after the consumption phase. Preliminary results showed that needle water content and colour were maintained until the trees were placed indoors (3 weeks after harvest), only xylem pressure potential decrease previously. The two species differed in particular on their colour characteristics. The *A. nordmanniana* were greener at harvest and turned less yellow than *P. abies* during the consumption phase. Moreover, *P. abies* had a poor needle retention compared to *A. nordmanniana*. In the same way, the colour characteristics also enabled to distinguish provenance: trees from Finistère were more yellow than others. In conclusion, it was clear that the critical phase was when trees were placed in drying conditions (consumption phase) and it might be possible to slow down this degradation. The next step will be to test improved process such as hydration, to maintain much longer the postharvest quality of Christmas trees.



NIBIO

NORWEGIAN INSTITUTE OF
BIOECONOMY RESEARCH

NIBIO - Norwegian Institute of Bioeconomy Research was established July 1 2015 as a merger between the Norwegian Institute for Agricultural and Environmental Research, the Norwegian Agricultural Economics Research Institute and Norwegian Forest and Landscape Institute.

The basis of bioeconomics is the utilisation and management of fresh photosynthesis, rather than a fossil economy based on preserved photosynthesis (oil). NIBIO is to become the leading national centre for development of knowledge in bioeconomics. The goal of the Institute is to contribute to food security, sustainable resource management, innovation and value creation through research and knowledge production within food, forestry and other biobased industries. The Institute will deliver research, managerial support and knowledge for use in national preparedness, as well as for businesses and the society at large.

NIBIO is owned by the Ministry of Agriculture and Food as an administrative agency with special authorization and its own board. The main office is located at Ås. The Institute has several regional divisions and a branch office in Oslo.

www.nibio.no