How your Christmas Tree Grower licensing fee benefits your industry.

Since 2012, the WSDA Christmas Tree Growers License program has provided:

- A total of $41,966 in support of ongoing disease and postharvest quality research. (See highlighted sections for details.)
- Continued communication between growers and WSDA/USDA in the event of a quarantine.
- And to maintain the delivery of Christmas trees from the grower to all living rooms around the world strong, healthy and pest free.

If you would like to learn more about the studies of WSU (Washington State University).

Please visit our webpage: agr.wa.gov ~ Plant ~ Insect ~ Christmas Tree Grower Licensing~Hot topics

Excerpt of projects supported with **WSDA Christmas Tree Licensing funding**.
Christmas Tree Licensing supported projects are highlighted in yellow.

Head researcher: Gary Chastagner, Ph.D., Professor of Plant Pathology,
WSU Research & Extension Center

Contact Professor Chastagner at **chastag@wsu.edu** or
WSDA Plant Services at **(360) 902-1874** for full report.

WSU Puyallup Christmas Tree Research

2016

Summary of Christmas Tree Projects

Development and use of genomic tools to improve firs for use as Christmas trees (Chastagner and Coats)

A five-year, $1.3 million grant from the National Institute of Food and Agriculture (NIFA) was awarded to Christmas tree researchers from Washington State University, North Carolina State University, University of California at Davis, Michigan State University, and Pennsylvania State University to identify and develop genetic markers to identify trees that have superior needle retention and resistance to Phytophthora root rot. The project began in October 2012.

Focusing on true firs (*Abies* spp.), the researchers involved in the NIFA project will build on the success of previous industry-supported research at WSU-Puyallup and North Carolina State Universities by involving the genomics groups at North Carolina State University and the University of California, Davis to find genetic markers to identify sources of trees that exhibit superior needle retention and are resistant to Phytophthora root rot.

Needle loss is one of the biggest concerns for families purchasing real Christmas trees. Decades of postharvest needle retention work at WSU has identified phenotypes of Douglas-fir, balsam, Canaan, Nordmann, and Turkish firs that have excellent needle retention as well as those that tend to exhibit extensive shedding within a few days of drying. Over the past 15 years, scion wood from tested trees has been used to establish about 3 acres of clonal holding blocks at WSU-Puyallup.

To identify genetic indicators, or markers, for needle retention, Fraser, balsam and Canaan fir trees were sampled in 2012 and 2013. The very small area at the base of each fir needle (abscission zone) was dissected from a branch from the current-year's growth. RNA was isolated from pooled abscission zones corresponding to trees with either very good or very poor post-harvest needle retention. The resulting RNA libraries were indexed separately by applying unique oligonucleotide barcodes, then pooled together and run on two lanes of an Illumina Hiseq 2000 high-throughput sequencing system. The sequences were assigned back to individual trees using the oligonucleotide barcodes. Analysis of this data is underway to identify not only possible genes involved in needle retention but gene expression patterns that diverge between the two populations. The next step is to target the regions with differences to develop a diagnostic assay that will predict whether a seedling will grow into a Christmas tree with good needle retention.

Similar techniques will be used to identify trees that are resistant to Phytophthora root rot (See below). This disease plagues growers in all regions where Christmas trees are grown and often limits where consumer-desirable species, such as noble and Fraser fir, can be grown. There is no effective control for this disease, so the best way to tackle the problem is to find resistant tree species and strains. Considerable work on Phytophthora root rot has been done at WSU and North Carolina State University.

Compared to the growth in the number of artificial trees used today, the actual number of real Christmas trees sold in the U.S. has remained relatively static for decades. To address the stalled market for real Christmas trees, the NIFA grant will also support Jeff Joireman at WSU, who will research consumer preferences. This work will be done in collaboration with the National Christmas Tree Association (NCTA). As a part of this project, we seek to further explore consumer preferences that may transcend the differences between species and ultimately genetically improve some, and perhaps eventually all species to become more consumer-friendly.

Postharvest

The role of ethylene in needle loss on cut Christmas trees (Chastagner, DeBauw, McLoughlin, and Mattheis).

Washington and Oregon Christmas tree growers supply approximately 40% of the total number of Christmas trees sold in the U.S. About 90% of these trees are shipped outside of the PNW to markets throughout the U.S. and exported to a number of foreign countries. Industry surveys have shown that needle loss is one of the top reasons consumers cite for dissatisfaction with fresh Christmas trees. Needle loss is a common problem with most species of conifers, including PNW-grown Douglas-fir, that are grown as Christmas trees. Considerable research has been conducted trying to identify factors that predispose cut trees to shed needles. As expected, early harvest and allowing trees to dry afterward increases needle loss problems. While delaying harvest helps reduce needle loss, this is difficult to do when growers must ship trees to retailers in other states who want to open their lots after Thanksgiving.

Recent research on balsam fir Christmas trees in eastern Canada has shown that exposure to concentrations as low as 10 ppm of ethylene will significantly accelerate postharvest abscission of balsam fir needles. Ethylene is a simple unsaturated hydrocarbon, which regulates many diverse metabolic and developmental processes in plants. The most studied process related to ethylene is its role in senescence and abscission on various plant tissues. Typically, ethylene is thought to increase prior to abscission and stimulate the activity of several enzymes, such as cellulase and pectinase. The enzymes have a variety of roles, which include weakening of cell walls, dissolution of middle lamella, and swelling of cells in the abscission zone.

When balsam fir branches were exposed to a continuous source of ethylene, abscission was greatly accelerated. It is unclear what role ethylene plays in the loss of needles from other conifer species, such as Douglas-fir, that are more widely grown in the PNW. It is also unclear how effective treatments such as 1-MCP that are commonly used to inhibit the effects of ethylene on other crops would be on reducing needle loss and improving tree quality. We are using a series of clonally-propagated balsam fir, Douglas-fir, Nordmann fir, Turkish fir, and Canaan fir trees that are currently maintained at WSU Puyallup to determine the role of ethylene in needle loss on these four additional species of trees. During the past 15 years we have been working with growers in a number of states to identify individual Christmas trees that are genetically predisposed to retain their needles. The clones of these trees at Puyallup have been subjected to numerous needle loss tests and represent a unique set of genotypes that range from clones that are predisposed to either shed or retain needles when detached branches are displayed dry. This includes clones of Nordmann and Turkish fir that have been tested as part of a 3-year 2010 WSDA SCBG supported project. We are also determining the potential of reducing needle loss and improving tree quality by investigating the effectiveness of 1-MCP in reducing the loss of needle on these species.

Postharvest moisture status and quality of trees displayed in tenon-type Christmas tree stands (Chastagner and McReynolds).

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, Fraser, and Nordmann fir 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.

Variation in postharvest needle retention characteristics of Turkish and Trojan fir populations from Turkey (Chastagner, Kurt, Frampton, Isik, and Landgren).

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 populations (Adapazarı-Akyazı, Bolu-Aladağ and Karabük-Keltepe) and two Trojan fir (*A. equi-trojani*) populations (Çanakkale-Çan and Balıkesir-Kazdağı) 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-Aladağ 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-Kazdağı 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 (Chastagner, Mcloughlin, and Riley).

In 2008, a replicated common garden field trail 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 (*A. balsamea*) and eight progeny collections of 'bracted' balsam fir (*A. balsamea* var. *phanerolepis)*. A single source of Fraser fir 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 postharvest needle retention, care needs to be taken when selecting seed sources in order to insure the best tree quality as well as profitability.

Root Disease

***Phytophthora* root rot component of genomic project -** Katie McKeever, WSU Plant Pathology Ph.D. Graduate Student

A survey of Phytophthora species causing root rot of Abies in U.S. Christmas tree farms (McKeever and Chastagner).

Multiple *Phytophthora* species are known to cause root rot (PRR) on true firs (*Abies spp.)*. Information about the community structures and habits of these regionally-variable pathogens has received little study

Insects and Other Pests

Risk of Adelgids surviving on cut Nordmann fir Christmas trees and boughs (Chastagner, Riley, and McLoughlin).

In Europe where Nordmann fir is widely grown for Christmas trees and boughs, the silver fir woolly adelgid [*Adelges* (*Dreyfusia*) *nordmannianae*] 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 20C 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.

Effectiveness of hot water dips in eliminating slugs on exported Christmas trees (Chastagner, DeBauw, and McLoughlin).

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

Cold Storage Facilities

Using funds from the WSDA Christmas Tree License Surcharge, a head house that was constructed in the early 1980's to conduct postharvest research on Christmas trees at WSU Puyallup was converted into a 400 square foot cold room. Because this space was recently infested by a colony of bats it is known as the "Bat House" by program employees. Improvements included close-cell insulation blown into the walls and ceiling, and the installations of a cooling system, lighting, and storage racks. Temperature in the room can be maintained from 29°F upward to ambient temperature. This room will be used for multiple projects ranging from cold storage trials for Christmas trees and bough material to storage of seedlings.