

Progress Report
Conifer Translational Genomics Network
Y1Q3 – April 1, 2008 to June 30, 2008

General Progress: For the third quarter of the CTGN (Conifer Translational Genomics Network), progress continued in all six objectives. Specific details are given below.

The first CTGN annual meeting took place June, 3-5 on the North Carolina State University campus in Raleigh, NC. In addition to team members, the meeting was attended by 13 additional invitees including members of the Scientific, Education, and Extension Advisory committees, as well as stakeholders, and USDA management staff.

Project Coordinator Andrew Bower has taken a position with the US Forest Service, and his last day was June 27. Recruitment is under way to fill a lab manager position to cover the running of the new DNA extraction facility, and an administrative person will be sought in the near future.

Objective 1: Validate previously discovered SNP by trait associations in Cooperative operational tree improvement populations.

University of Florida – CFGRP funded MS student Patricio Munoz continues with phenotyping of the BC1 population

Work has begun to learn and validate Jessica Li's Bayesian program for SNP analysis and missing SNP imputation. In conjunction with that effort, SNP data from a structured population is being analyzed with various methodologies to insure that appropriate SNP effects are estimated and declared significant.

Texas A&M University - no additional progress during this reporting period

Oregon State University – A draft has been completed of Module 1 of the Tree Genome Simulator (software and documentation) and was demonstrated at the CTGN Annual meeting in Raleigh, NC. This program will simulate quantitative trait phenotypic data and corresponding genetic marker (SNP) haplotype data that can then be used to explore alternative sampling and analytical strategies for discovering and quantifying SNP/phenotype associations in tree breeding populations.

A daylong CTGN research and extension workshop was held June 25, 2008 in Aurora, OR. This workshop was organized by Glenn Howe, Nicholas Wheeler, and Dave Harry and had 24 attendees

North Carolina State University – Phenotyping continues on the Genetics X Culture study that will be genotyped in the coming months. Densitometry is completed for 3 of the 5 reps and

should be completed for all 5 of the reps in the next 3 weeks. Samples for genotyping will be collected in the next few weeks.

In the Atlantic Coastal Elite Population (ACE), 5-month rust assessments have been completed and analyzed. This has turned out to be a great data set with great potential for CTGN. Progeny from 76 crosses among the best genotypes in the Atlantic Coastal Plain were screened for fusiform rust resistance at the USDA Forest Service Resistance Screening Center in Asheville. At 5 months, the average infection was 48%; the family-mean heritability was 0.95; and families ranged from a low of 8% galled to a high of 84%. The gall-free trees will be hedged to promote cuttings for subsequent clonal testing.

Analyses continue on the Lower Gulf Elite Population, and manuscript preparation on the genetic analyses for sawtimber potential continue.

We have identified specific tests in our database that will be sampled for CTGN. We are focusing initial efforts on the best quality trials (low CV's, high survival and growth, high heritabilities for growth) for our initial genotyping.

University of California at Davis – The Eppendorf epMotion 5075 LH was the chosen platform for automated, high-throughput DNA extraction, using the Invitrogen ChargeSwitch plant kit. Homogenization of samples will be done using the SPEX Sample Prep Genogrinder 2000. These instruments have been purchased and are in place in the lab and personnel are being trained and protocols optimized.

Discussions regarding barcoding of samples and methods of sample collection in the field took place by phone and e-mail among team members. Protocols for sampling of materials in the field and for printing barcodes for labeling of sample vials were then developed and distributed to team members for review and implementation.

Recruitment is under way for a Plant Genome Lab Manager to continue the scientific aspects of the CTGN Project Coordinator position vacated by Andrew Bower. This position will entail both management of the lab and performing DNA extraction on samples received from team members. Administrative duties will temporarily fall back to the PD and co-PI's, with the intention of filling a part-time Project Coordinator position to handle administrative duties starting in January of 2009.

Objective 2: Develop and economically evaluate new methods incorporating marker-assisted selection into conifer tree breeding programs.

An Excel based simulation model to do economic analysis for alternative breeding strategies has been developed through a contract with Dr. James Richardson in the Agricultural Economics Department at Texas A&M University. The program is intended to contribute to objectives 5 and 6 when the user interface is complete as it illustrates how breeding cycles work and has

graphic comparison showing relative returns and risks on various options. The model was demonstrated at the CTGN annual meeting in Raleigh, NC.

Objective 3: Develop databases (TreeGenes) and web-based tools to facilitate all aspects of the CTGN project.

Ben Figueroa was hired to fill the developer position and started on May 15th. Recruitment for the programmer 1 position is still under way. Ben has been working on development of the resequencing database for future use by CTGN.

The Plone website is now up and fully operational. The information that was previous available on the Wiki site is now available on the Plone site. Team members have started utilizing the site for posting documents and many of the presentations given at the annual meeting in Raleigh, NC are now posted here.

The CTGN website is continually updated as needed, and pipelines for handling the large databases that will be generated by this project continue to be developed, building off of the progress made by earlier projects (ADEPT2).

Objective 4: Develop an international genetic stock center for conifers.

University of Georgia - The project has already suffered its first turnover in personnel. After 12 weeks on the job, our new technician, Ms. Holley Langille, had to resign for personal reasons. The search has been re-opened, and Dr. Lorenz is currently interviewing applicants for the position. We hope to have the position filled by the end of June.

Prior to her departure, Ms. Langille completed preparation of total RNA samples for *Pinus taeda*, *P. palustris*, *Podocarpus macrophylla*, *Cephalotaxus harringtonia*, and *Sciadopitys verticillata*. These samples have been shipped to the JGI, where the *P. taeda* sample is currently being used to prepare normalized and non-normalized cDNAs. The other four RNA samples will be used to prepare normalized cDNAs as soon as the *P. taeda* samples are completed.

Douglas-fir - Seed for full-sib Douglas-fir families with high graft compatibility were sown in May. Germination was good. More than 1,400 seedlings are being grown in the greenhouse in Corvallis. Scion from important Douglas-fir germplasm will be grafted onto the rootstock, possibly beginning next winter.

Loblolly pine – 113 clones of the Base2 and QTL2 pedigrees were successfully grafted in 2008, and have been planted into the Harrison Experimental Forest (HEF) (Saucier, MS). Approximately 350 clones from the 2007 Base2 and QTL2 pedigrees have been planted in a clonal archive. The planting is a randomized complete block with two reps of two-tree clonal plots, with trees planted on a 15 x 25 foot spacing. Clones having more than four ramets are being planted in an overflow area on the Erambert/Black reek Seed Ochards (U.S. Forest Service, Region 8) near Brooklyn, MS.

Seeds for rootstocks for FY09 grafting have germinated successfully and are growing in the HEF Greenhouse. Approximately 1000 rootstocks will be available for grafting in 2009.

Objective 5: Develop an education plan for undergraduate and graduate curriculum in genomics-based breeding for forest trees.

Draft outlines and collections of PowerPoint slides for Modules 4-6 has been completed, and draft outlines for Modules 1-8 are now posted on the Plone website.

Dave Harry and Nick Wheeler each prepared 2-page summaries for each workshop module, containing sections for Introduction, Key Messages, Outcomes, and Outline. These documents along with the workshop syllabus were assembled into a comprehensive handout for the CTGN 2008 annual meeting.

A comprehensive introductory presentation covering both Education and Extension activities (obj 5 & 6) was presented at the CTGN 2008 annual meeting.

Dave Harry and Nick Wheeler worked with Education and Extension evaluator Michael Coe to develop survey instruments for the education and extension activities. These materials have been approved by the Institutional Review Board (IRB) at UC Davis – a mandatory step prior to distributing survey material. Survey instruments have been developed for workshop participants as well as extension staff, and other generic survey materials have been developed for participants of future education or extension events.

Opportunities to interface CTGN interests with Christmas tree growers were discussed with OSU Extension Specialist Chal Lingren on April 16.

Discussions are under way on how to provide coop staff (and certain coop members) information being developed from our educational workshop. We are exploring ways to either facilitate these people attending the workshop itself, or alternatively, putting on a 2- to 3-day workshop with content specifically designed for coop participants.

Objective 6: Develop an extension plan for continuing education in genomics-based breeding for practicing tree breeders and forest tree gene resource managers. Develop plans for extension evaluation of all activities.

Michael Coe presented the concepts and approaches to extension and education evaluation at the annual CTGN meeting in Raleigh, and project directors were engaged in filling out questionnaires for baseline data gathering. The presentations given by Coe were well received by staff and advisory committee members as well as by CSREES staff.

Project personnel have made a number of presentations to a wide range of audiences highlighting the proposed activities of the CTGN or essence of association genetics in plants/trees (Table 1).

Table 1. Extension, education and outreach presentations of the Conifer Translational Genomics Network during quarter 3.

Activity	Presenter/ Author	Title	Date & Location	Venue & Audience
Oral Presentation	Neale, D.B.	Population, quantitative and comparative genomics of adaptation in forest trees	April Vancouver, B.C.	University of British Columbia forest sciences seminar
Lecture	Whetten, R.	Molecular markers and their applications in forest species	April 10, Concepcion, Chile	Curso Internacional de Genetica y Biotecnologia Forestal
Lecture	Whetten, R.	Functional genomics in forest species	April 10, Concepcion, Chile	Curso Internacional de Genetica y Biotecnologia Forestal
Oral Presentation	Byram, T.	Conifer Translational Genomics Network: The CAP Grant – Bringing markers to bear on applied tree breeding program.	May 13-14, College Station, TX	Western Gulf Forest Tree Improvement Program contact meeting
Oral Presentation	McKeand, S.	Update on CAP grant, Conifer Translational Genomics Network	May 28, Atlanta, GA	52 nd Advisory committee meeting of the NC State University Cooperative Tree Improvement Program, 41 attendees
Oral Presentation	Howe, G.T. et al.	Genetics of Douglas-fir wood stiffness: coustic tools, bending stiffness, and candidate gene markers	May 28, Vancouver, WA	The Science of Wood Quality: Workshop jointly sponsored by PNWTIRC, NWTIC, SMC, and WFCA. 75 attendees
Oral Presentation	Huber, D.	CTGN and its direct applicability to the CFGRP		Cooperative Forest Genetics Research Program annual meeting
Oral Presentation	Munoz, P.	BC1 phenotyping results		Cooperative Forest Genetics Research Program annual meeting
Web Seminar	Neale, D.	GoldenGate and Infinium SNP Genotyping for association studies in trees	June 20, On-line	Public
Oral Presentation	Howe, Wheeler and Harry	CTGN research and extension workshop	June 25, Aurora OR	PNWTIRC members and staff, 24 attendees