Progress Report
Conifer Translational Genomics Network
Y1Q2 – January 1, 2008 to March 31, 2008

General Progress: The second quarter of the CTGN (Conifer Translational Genomics Network) showed progress in all six objectives. Specific details are given below. The first annual meeting of the team will be taking place June, 3-5 on the North Carolina State University campus in Raleigh, NC. Many of the team members met informally at the International Plant and Animal Genome conference in San Diego, CA in January, where they collectively presented over a dozen posters and oral presentations describing the science that is the basis for the CTGN. Sub-contract awards to collaborating institutions have been established with details being worked out by individual institutions. Andrew Bower started as project coordinator on January 7, 2008.

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

University of Florida – Phenotypic measurements are being taken weekly on the hybrid (slash pine X loblolly pine) X slash pine trial at High Springs, FL. The measurements began February 23rd and consist of height and status of the seedling. Frequent measurement early and late in the growing season will ensure that the timing for growth initiation and cessation are captured. During the middle of the growing season, measurements will be at 4-week intervals.

Funding for the University of Florida subcontract began on 3/5/2008

Texas A&M University - The acoustic sampler being used for the preliminary field trial of the Time of Flight method for measuring wood stiffness was broken and has not been able to be repaired. Daniel Chmura did not accept the position offered and another candidate has not been identified.

Oregon State University - Recruitment for the quantitative geneticist post-doctoral research associate at OSU is completed. The search committee (Glenn Howe, Dave Harry, Marilyn Cherry, and Terrance Ye) chose Jianbin Yu, who will start on April 1, 2008.

North Carolina State University – Research assistant Josh Steiger has been learning the intricacies of X-ray densitometry and is now proficient at preparing and processing cores for the QMS Densitometer as well as data editing after the scanning. Scanning is approximately 40% complete on the Genetics X Culture study that will be genotyped in the coming months.

Once shoot and foliage elongation has progressed to the appropriate stage (May - June, depending on the location of the trees), tissue will be collected for DNA extraction.

Data collection is continuing on the Lower Gulf Elite Population. Breeding values have been

determined for growth and numerous stem form traits (sweep; branch angle, frequency, size; forking; ramicorn branching; stem and branch sinuosity; and sawlog potential). TreeSonic measurements for wood stiffness are underway.

NIR machining and calibration continues, with full NIR capabilities for scanning cores anticipated within 6 months.

Jaime Zapata will start as a PhD student in August. Jaime currently works for Arauco in Concepcion, Chile and will bring several years of genetics and forestry experience to our program.

University of California at Davis – Planning is complete for SNP genotyping to be completed at the DNA Technologies Core facility at UC Davis. The full set of loblolly pine selections from the Texas A & M and North Carolina State University breeding programs will be genotyped for a total of 17,968 SNP's, which is in excess of 10X the number put forward in the project proposal. This will be done in two parts. The first part will be a 7,600 SNP Illumina Infinium chip that has been designed under the ADEPT 2 project and is being tested for associations with phenotypes in 3 association populations. The second part will be on an additional Infinium chip to be designed and manufactured that will contain 10,368 SNP's. Genotyping is anticipated to take place during the first and second quarter of year two.

Needle and seed tissue was sent out to five different companies to test their automated DNA extraction equiptment and protocols (Qiagen, Eppendorf, Agencourt, Corbett Robotics, and AutoGen). Results were highly variable and follow-up tests were necessary to allow the choice of an effective and efficient platform for use in the DNA extraction lab. A decision on which platform to purchase will be made in April. The SPEX Sample Prep Geno/Grinder 2000 bead mill was selected as the method of choice for high-throughput tissue grinding and purchasing of this machine has been initiated. A target date of early June has been set for the lab to be operational with sample preparation and DNA extraction anticipated to take place during the fourth quarter of year one, and the second quarter of year 2.

Discussions have taken place among co-PI's and the project coordinator to develop a protocol for field sampling of materials to be genotyped. The tentative protocol calls for needles to be sampled and placed into plastic vials with a dessicant to dry the tissue for transport and storage until DNA is extracted. Sample vials will be labeled at the time of collection and then barcoded for further tracking through the process of transport to Davis and DNA extraction. Approximately 5000 samples will be collected this summer from the North Carolina State University and Texas A & M populations.

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

A draft flow chart and spreadsheet template for an economic simulation package (Simetar) to be implemented in Excel has been created. This package will eventually allow side-by-side comparisons of several different breeding strategies and sensitivity analysis to allow the identification of breeding activities with the greatest economic impact, and incorporate presentation graphics to allow this information to be easily communicated to decision makers. The Simetar model was discussed with Dr. James Richardson, a professor in Agricultural Economics and the creator of the model and he has agreed to assist with the completion of building the model and ensuring that all comparisons are done according to accepted economic standards and that the final product has a professional appearance.

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

Lead programmer Jennifer Lee left Davis for a database administrator position with NCBI in Bethesda, MD. Jill Wegrzyn has assumed responsibility as lead programmer and is in Davis part time now and will be moving to Davis permanently later this summer. Alex Voong was hired as a programmer (grade I) to help with website and database management. Recruitment is underway for a database manager and an additional mid-grade programmer. A significant amount of work was done to design and build the Plone platform for data and project management for CTGN. The Plone site went live in March, and file uploading began almost immediately. While there are still some kinks to work out, it appears as though it will be a highly efficient way to network and share files among team members, as well as to plan and organize progress on all 6 main objectives and their sub-objectives

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.

Douglas-fir - Seed for full-sib Douglas-fir families that have high graft compatibility (95-98% compatible) have been obtained and been placed into cold stratification. Seed will be removed from stratification in April and sown and rootstock seedlings will be grown for two years before being ready to graft.

Loblolly pine – Grafting of the Base and QTL loblolly pine reference pedigrees was completed in FY08. An inventory of surviving ramets for these populations will be available for the June 30 report. Arranging for additional space to establish the grafted ramets of the loblolly pine component of the stock center was initiated for both the Harrison Experimental Forest near Saucier, MS and the Erambert/Black Creek Seed Orchards near Brooklyn, MS. Seeds for root stocks for FY09 grafting were put into stratification for spring 2008 sowing and growing.

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

PI's met Feb. 5-6, 2008 (Harry, Wheeler, Howe) on the Oregon State University campus to discuss and plan workshop activities. A draft syllabus and a task work schedule for developing workshop materials were developed for distribution to CTGN team members to receive feedback prior to the annual meeting in June.

PI's visited the Neale lab group in Davis, CA March 11-13, 2008 to gain clarity and understanding of the TreeGenes bioinformatics pipeline so as to represent it properly and accurately in the workshop.

Various materials have been gathered for use in the workshops in presentations, printed material, and suggested references. These materials include: lecture presentations from UC Davis (Neale, Dubcovsky, and Famula), assorted images, and recent texts on genetics, population genetics, genomics and bioinformatics.

Curriculum outlines and draft powerpoint presentations for Modules 1 (Workshop introduction and Basic principles in population and quantitative genetics), 2 (Introduction to conventional tree breeding), 3 (Genetic Markers), 4 (Molecular population genetics), 7 (MAS: parameter estimation), and 8 (Genetic gain and value estimation) have been completed. Progress has been made on modules 5 (QTL mapping) and 6 (Association genetics) with completion planned for late April.

The full workshop syllabus will be presented to CTGN team members, with an opportunity to review course materials and presentations, at the annual meeting in Raleigh in June.

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.

The contract proposal for Michael Coe, our chosen extension evaluation expert was acted upon by UC Davis officials mid-quarter, requiring additional feedback to accommodate sole-source (non-competitive) contracting. A UD Davis contract was ultimately sent to Dr. Coe. The signed return copy of the contract is now in process through the UC Davis administration and should be in place shortly.

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 2.

Activity	Presenter/ Author	Title	Date & Location	Venue & Audience
Poster Presentation	Neale et al.	Conifer Translational Genomics Network	January 12-16, 2008 San Diego, CA	International Plant & Animal Genome XVI. Over 1000 attendees.
Oral Presentation	Wheeler, N.	The role of genomics in tree breeding	January 31, 2008, Vernon, B.C. Canada	BC Tree Breeders annual meeting. 16 attendees comprised of tree breeders and managers.
Oral Presentation	Neale, D. B	From Genes to Trees: Genomics, tree physiology and forest ecology	February 2008 Victoria, Canada	Forest Biology Symposium. University of Victoria
Oral Presentation	Neale, D. B	Association studies with molecular markers.	February 2008 Davis, CA	Breeding with molecular markers. UCDavis Extension
Oral Presentation	Neale, D. B	Genomics to Tree Breeding and Forest Health	February 2008 Coeur d''Alene, ID	Inland Empire Tree Improvement Cooperative Annual Meeting
Oral Presentation	Howe, G.T./Howe, G., Strauss, S., and Jayawickrama, K.	Overview of CAFS-related Research at Oregon State University	February 20-21, 2008. Portland, OR	Annual meeting of the NSF Center fore Advanced Forestry Systems (CAFS). 65 Participants.
Oral Presentation	Neale, D.B.	Tree Breeding: Conquering high heterozygosity and long generation times	March 17, 2008 St. Paul, MN	Plant Breeding in the 21st Century: Challenges and Opportunities Across the Plant Sciences. XXX Attendees
Oral Presentation	Jermstad. K.D.. /Jermstad, K.D. and Neale D.B.	Genetic Mapping of Genes Controlling Slow-Rusting Resistance and Major Gene Resistance in Sugar Pine (Pinus lambertiana)	March 26-28, 2008. Walnut Creek, CA	3rd Annual DOE Joint Genome Institute Users Meeting. Approx. 350 attendees. Mostly researchers on biofuels